publications of the INSTITUTE of MARINE SCIENCE

EDITOR'S NOTE

This is a special issue entirely devoted to papers summarizing part of the extensive unpublished reports on oysters and their relationship to the oil industry prepared from 1947 to 1960 as part of projects 9 and 23 of the Texas A and M Research Foundation. As part of its responsibility for dissemination of knowledge in the marine sciences, the Publications of the Institute of Marine Science joins the Texas A and M Research Foundation in publishing at joint expense summaries of this important work.

Volume 7 For 1961

Published by the
Institute of Marine Science
The University of Texas
Port Aransas, Texas
July, 1962

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Publications of the Institute of Marine Science
Volume 7

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The *Publications of the Institute of Marine Science* is printed annually by The University of Texas and includes papers of basic or regional importance by Gulf workers or on Gulf waters in Marine Science and related fields of Bacteriology, Botany, Chemistry, Geology, Meteorology, Physics, and Zoology. Papers are read by three referees; papers by the editors are processed by the Chairman of the Budget Council.

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Studies on Oysters in Relation to the Oil Industry

JOHN G. MACKIN AND SEWELL H. HOPKINS

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Preface

In the Louisiana parishes of Plaquemines, Jefferson, Lafourche, and Terrebonne there is a large commercial production of oysters, *Crassostrea virginica* (Gmelin), on bottoms leased from the state by individuals. The oysters on these leases are the private property of the leaseholder, by law. In 1946 and 1947 a number of the leaseholders filed claims, some of which later became law suits, asking compensation for the claimed financial losses. These claims were based on abnormally high mortality of oysters on their leases, allegedly caused by operations of certain oil and sulphur companies. Several of the oil companies asked the Texas A & M Research Foundation, an independent non-profit organization with headquarters on the campus of the Agricultural and Mechanical College of Texas, to undertake at their expense an impartial scientific investigation of the extent and cause of oyster mortality in Louisiana and Texas. From the beginning, scientists of The University of Texas, Texas Christian University, and Louisiana State University, as well as the A & M College of Texas, were enlisted in this large cooperative research project, which ultimately cost over 2 million dollars. The researches directed by the senior scientists served to train many young chemists and marine biologists, and became widely known in the region as "projects 9 and 23." Most of the findings have not heretofore been published.

The original reports of Projects 9 and 23 that bear on this question are listed under "Literature Cited" and indicated by an asterisk. A few of these reports were later published in whole or in part, but most are in manuscript form only. A set of "processed" (mimeographed or lithoprinted) reports, including most of those cited here, has been sent to each of the institutional libraries named below. Extra copies of some of these reports are still available and will be sent to any institution or individual who will agree to pay postage and packing costs. The list of available reports will be mailed by either of the authors on request. Some additional reports that are not on the list have been deposited in the libraries of Virginia Fisheries Laboratory and of the United States Bureau of Commercial Fisheries, Annapolis, Maryland. Copies of these and all other reports may be obtained on 30-day loan from the authors.

The following 28 libraries were each furnished with a set of the reports that were available in sufficient number to permit distribution.

Agricultural and Mechanical College of Texas, Oceanography and Meteorology Library, College Station, Texas.
Bingham Oceanographic Laboratory, Yale University, New Haven, Conn.
Biological Station, Fisheries Research Board of Canada, St. Andrews, New Brunswick, Canada.
Chesapeake Bay Institute, Johns Hopkins University, Baltimore, Md.
Chesapeake Biological Laboratory, Solomons, Md.
Duke University Marine Laboratory, Beaufort, N.C.
Florida State Board of Conservation, Marine Laboratory, St. Petersburg, Fla.
Hawaii Marine Laboratory, University of Hawaii, Honolulu, Hawaii.
Institute of Fisheries Research, University of North Carolina, Morehead City, N.C.
Institute of Marine Science, University of Texas, Port Aransas, Texas.
Louisiana Department of Wild Life and Fisheries, New Orleans, La.
Narragansett Marine Laboratory, University of Rhode Island, Kingston, R.I.

Oceanographic Institute, Florida State University, Tallahassee, Fla.
Pacific Biological Station, Fisheries Research Board of Canada, Nanaimo, British Columbia, Canada.
Pacific Marine Station, College of the Pacific, Dillon Beach, Calif.
Rutgers University, Department of Zoology, New Brunswick, N.J.
Scripps Institution of Oceanography, La Jolla, Calif.
Texas Game and Fish Commission Marine Laboratory, Rockport, Texas.
U.S. Bureau of Commercial Fisheries, Gulf Fishery Investigation, Galveston, Texas.
U.S. Bureau of Commercial Fisheries Biological Laboratory, Gulf Breeze, Fla.
U.S. Bureau of Commercial Fisheries Biological Laboratory, Milford, Conn.
U.S. Fish and Wildlife Service Library, Woods Hole, Mass.
University of British Columbia Institute of Oceanography, Vancouver, B.C., Canada.
University of Delaware Marine Laboratory, Lewes, Del.
University of Georgia Marine Institute, Sapelo Island, Ga.
University of Oregon Library, Eugene, Oregon.
University of Washington Fisheries-Oceanography Library, Seattle, Wash.
Virginia Fisheries Laboratory, Gloucester Point, Va.

List of Unpublished Reports Deposited in Libraries

Boswell, J. L. March 20, 1950. Report on experiments to determine the effects of a surface film of crude oil on the absorption of atmospheric oxygen by water. 6 pp. Mimeo.

Boswell, J. L. July 6, 1950. The effect of crude oil on oysters (Gowanloch method). 35 pp. Mimeo.

Brown, S. O. May 31, 1950. Microbial oxidation of crude oils and water soluble fractions of crude oil as shown by the B.O.D. method. 2 pp. Mimeo.

Brown, S. O. and Virginia Van Horn. May 31, 1950. Aerobic and anaerobic oxidation of crude oils by microorganisms from Louisiana bay-bottom muds. 4 pp. Mimeo.

Brown, S. O. and Bobby L. Reid. May 31, 1950. Report on experiments to test the diffusion of oxygen through a surface layer of oil. 5 pp. Mimeo.

Brown, S. O., Virginia Van Horn and Bobby L. Reid. May 31, 1950. Decomposition of organic compounds by marine microorganisms. 11 pp. Mimeo.

Cauthron, F. F., J. G. Mackin and S. H. Hopkins. December 7, 1951. Experiments to test the effects of shell-drilling and mud-injection upon oysters, 1948. 45 pp. Mimeo.

Hewatt, W. G. July 26, 1950. Studies on dissolved oxygen content and hydrogen ion concentration in the waters of Barataria Bay, Louisiana, 1945–1947. 11 pp. Mimeo.

Hewatt, W. G. (No date: circa 1951). An oyster feeding experiment. 28 pp. Mimeo.

Hewatt, W. G. June 15, 1951. Salinity studies in Louisiana coastal embayments west of the Mississippi River. 156 pp. Mimeo.

Hewatt, W. G. May 22, 1953. Ecological studies on salt marsh ponds in southern Louisiana. 38 pp. Multilith.

Hewatt, W. G. June 15, 1953. An oyster mortality study in Lower Barataria Bay, Louisiana. 11 pp. Mimeo.

Hewatt, W. G. August 12, 1953. Climatological and hydrological records on the Barataria Bay area, Louisiana. Part II. 44 pp. Multilith.

Hopkins, S. H. November 11, 1948. Notes for a working hypothesis of the causes of oyster mortality in Louisiana and Texas. 9 pp. Mimeo.

Hopkins, S. H. February 1, 1950. History of some oyster mortalities reported in foreign countries. 21 pp. Mimeo.

Hopkins, S. H. February, 1950. A brief list of references on oyster mortality in some foreign countries. 5 pp. Mimeo.

Hopkins, S. H. July 20, 1950. The inter-relationship of weight, volume, and linear measurements of oysters and the number of oysters per Louisiana sack measure. 14 pp. Mimeo.

Jensen, F. W. and W. M. Potts. January 16, 1953. Mud samples and analyses. Part II. Volatile hydrocarbons, carbon tetrachloride extracts, and unsaponifiable residues in muds. 58 pp. Mimeo.

Lunz, G. R., Jr. May 2, 1950. The effect of bleedwater and of water extracts of crude oil on the pumping rate of oysters. 107 pp. Mimeo.

Mackin, J. G. April 11, 1950. Report on a study of the effect of application of crude petroleum on salt grass, *Distichlis spicata* (L.) Greene. 8 pp. Mimeo.

Mackin, J. G. April 12, 1950. A comparison of the effect of application of crude petroleum to marsh plants and to oysters. 4 pp. Mimeo.

Mackin, J. G. April 11, 1950. A report on three experiments to study the effect of oil bleedwater on oysters under aquarium conditions. 5 pp. Mimeo.

Mackin, J. G. April 19, 1950. The effect of crude petroleum on oysters heavily infested with Polydora, Cliona, and Martesia. 5 pp. Mimeo.

Mackin, J. G., Billy Welch and Charles Kent. April 10, 1950. A study of mortality of oysters of the Buras area of Louisiana. 49 pp. Mimeo.

Mackin, J. G. and D. A. Wray. February 1, 1949. A study of mortality and mortality-producing agencies in Barataria Bay, Louisiana. 73 pp. Mimeo.

Mackin, J. G. and D. A. Wray. August 8, 1950. Report on the second study of mortality of oysters in Barataria Bay, Louisiana, and adjacent areas. Part I. 49 pp. Mimeo.

Mackin, J. G. and D. A. Wray. August 9, 1952. Report on the second study of mortality of oysters in Barataria Bay, Louisiana, and adjacent areas. Part II. 59 pp. Mimeo.

Menzel, R. W. July 27, 1950. Report on oyster studies in Caillou Island oil field, Terrebonne Parish, Louisiana. 116 pp. Mimeo.

Menzel, R. W. August 9, 1950. Report on oyster studies in Lake Felicity and Bayou Bas Bleu, Terrebonne Parish, Louisiana. 70 pp. Mimeo.

Menzel, R. W. August 12, 1950. Report of oyster investigation at the Dog Lake oil field, Terrebonne Parish, Louisiana. 13 pp. Mimeo.

Menzel, R. W. January 6, 1951. Report on oyster studies at Lake Pelto oil field, Terrebonne Parish, Louisiana. 20 pp. Mimeo.

Menzel, R. W. and S. H. Hopkins. September 11, 1951. Report on experiments to test the effects of oil well brine or "bleedwater" on oysters at Lake Barre oil field. Volume II. 126 pp. Mimeo.

Menzel, R. W. and S. H. Hopkins. July 21, 1951. Report on commercial-scale oyster planting experiments in Bayou Bas Bleu and in Bay Sainte Elaine oil field. 218 pp. Mimeo.

Menzel, R. W. and S. H. Hopkins. August 3, 1953. Report on oyster experiments at Bay Sainte Elaine oil field. 208 pp. Multilith.

Menzel, R. W. and S. H. Hopkins. July 24, 1954. Studies on oyster predators in Terrebonne Parish, Louisiana. 156 pp. Multilith.

Prokop, J. F. May 29, 1950. Report on a study of the microbial decomposition of crude oil. 81 pp. Mimeo.

Prokop, J. F. August 11, 1950. Infection and culture procedures employed in the study of *Dermocystidium marinum*. 25 pp. Mimeo.

Seiling, Fred W. June 26, 1951. Experiments on the effects of seismographic exploration on oysters in the Barataria Bay region, 1949–1950. 59 pp. Mimeo.

Seiling, Fred W. April 20, 1953. Experiments on the effects of seismographic exploration on oysters. 11 pp. Mimeo.

Organization and Acknowledgments, Projects 9 and 23

Project 9 began February 1, 1947, and ended May 31, 1950. Project 23 began June 1, 1950, and was still continuing in 1960. Project 9 was a contract research project originating in the Department of Biology, Agricultural and Mechanical College of Texas, and sponsored by the Texas Company (now Texaco, Inc.), Humble Oil and Refining Company, The California Company, Tide Water Associated Oil Company (now Tidewater Oil Company), Phillips Petroleum Company, and Shell Oil Company. The same six companies plus Gulf Refining Company became joint donors of Project 23, which originated in the Department of Oceanography and Meteorology, A & M College of Texas.

The original contract specified that the Foundation should conduct to conclusion a thorough, scientific and impartial research to determine whether or not an abnormal oyster mortality was occurring or had occurred during the past 12 months in Louisiana or Texas waters near operations of the sponsoring companies, the degree of abnormal mortality, if any, its cause, and, if connected with oil company operations, exactly how it was connected.

As outlined in a memorandum by Dr. A. A. Jakkula (1947), Executive Director of the Texas A & M Research Foundation, on July 15, 1947, Project 9 had the following research divisions: Bacteriology under Dr. S. O. Brown of the A & M College of Texas, Chemistry under Dr. F. W. Jensen of A & M, Ecology under Dr. Willis G. Hewatt of Texas Christian University, Field Studies on Oysters under Dr. J. G. Mackin of A & M, Geology under Professor S. A. Lynch of A & M, Library under Dr. C. C. Doak of A & M, Parasitology under Dr. H. J. Bennett of Louisiana State University, Pathology and Histology under Dr. S. O. Brown of A & M, and Physiology under Dr. E. J. Lund of The University of Texas. The Parasitology division did its work at Grand Isle and at the Baton Rouge campus of L.S.U., and Physiology was centered at the Institute of Marine Science of The University of Texas, Port Aransas. Dr. Hewatt of the Department of Biology and Geology, T.C.U., continued a study of the hydrobiology of Barataria Bay that he had been engaged in before Project 9 started. Dr. S. H. Hopkins was Director of Project 9, Dr. J. G. Mackin was Director of Field Work and Assistant Director of Project, and Dr. C. C. Doak was Coordinator. The consultants listed in the 1947 outline were Dr. Thurlow C. Nelson of Rutgers University, Dr. Herbert F. Prytherch of the U. S. Fish and Wildlife Service, Dr. C. R. Elsey of British Columbia Packers, Dr. Claude E. ZoBell of Scripps Institution of Oceanography, and Mr. H. A. Marmer of the U. S. Coast and Geodetic Survey. In 1948 Dr. P. Korringa of Bergen op Zoom, the chief

shellfish biologist of Holland, was added to the list of consultants. All of these men inspected work in progress in the laboratory and in the field and gave valuable advice based on first-hand study of the procedures of Foundation workers.

A revised plan of organization and a personnel chart for Project 9 were prepared by Dr. Jakkula on July 3, 1948. The chart is shown in Fig. 1. The research divisions were now listed as Bacteriology, Biological Technique, Chemistry, Ecology and Hydrobiology, Geology and Hydrography, Oyster Biology, Parasitology, and Physiology. Ecology and Hydrobiology was headed by Dr. Hewatt at Texas Christian University, Oyster Biology by Dr. Mackin at Grand Isle, Parasitology by Dr. Bennett at Louisiana State University, and Physiology by Dr. Lund at Port Aransas (Institute of Marine Science). All other divisions had headquarters at the A & M College of Texas, under Dr. Brown (Bacteriology and Biological Technique), Dr. Jensen (Chemistry), and Professor Lynch (Geology and Hydrography). The duties and activities of each research division were outlined in a mimeographed memorandum of the same date. Under Chemistry 22 different phases of research were listed, under Oyster Biology 26, and under Physiology 9.

Still other studies were added later. Some of these were conducted by persons at other institutions. Special bacteriological studies were carried out in the laboratory of Dr. Claude E. ZoBell at Scripps Institution of Oceanography, La Jolla, California. Physiological experiments were run at Bears Bluff Laboratories, Wadmalaw Island, South Carolina, under the direction of Mr. G. Robert Lunz, Jr. Mr. Fred Sieling of the Maryland Department of Research and Education directed a special field study of the effects of seismographic exploration on oysters at two localities in Louisiana. An annotated bibliography of oysters (794 pages) was compiled by Mr. J. L. Baughman of the Texas Game and Fish Commission and published by the Foundation.

Later on in the development of Project 9 there were further changes in the actual working of the organization. Most of these changes tended toward centering more of the work at Grand Isle, under the direct supervision of Dr. Mackin. During the most active period in 1947 and 1948 over 90 persons were employed on the project, and there were still 43 employees at the end.

As the contract indicated, the possible effects of oil operations and the improvement of commercial oyster-growing were the phases of Project 9 given priority at the beginning. However, studies of parasites and disease had been mentioned in the tentative plans for the investigation proposed on February 18, 1947. Divisions of Bacteriology and Parasitology were included in the original organization. By the summer of 1948 it had become certain that a disease was causing a constant mortality of oysters during the warm months in Louisiana waters west of the Mississippi River. It was increasingly evident that this disease was harmful to oyster production. The drilling operations, oil losses, and bleedwater effluents which had been pointed out by local oystermen began to look very doubtful as causes of significant mortality. The protozoan *Nematopsis* and the boring worm *Polydora* were investigated as possible causes of disease in 1947 and 1948, but the organism that caused most of the mortality remained unknown. Bacteriological studies gave no promising leads. Studies of diseased tissues and blood of oysters in the summer and fall of 1948 showed that a yeast-like organism was present in most gapers. These were studied in live smears. Sectioning of diseased oysters by Mackin in December, 1948 revealed a tissue-damaging organism of unknown nature. After a long literature search by Hopkins it was tentatively identified on August 2, 1949, as a new

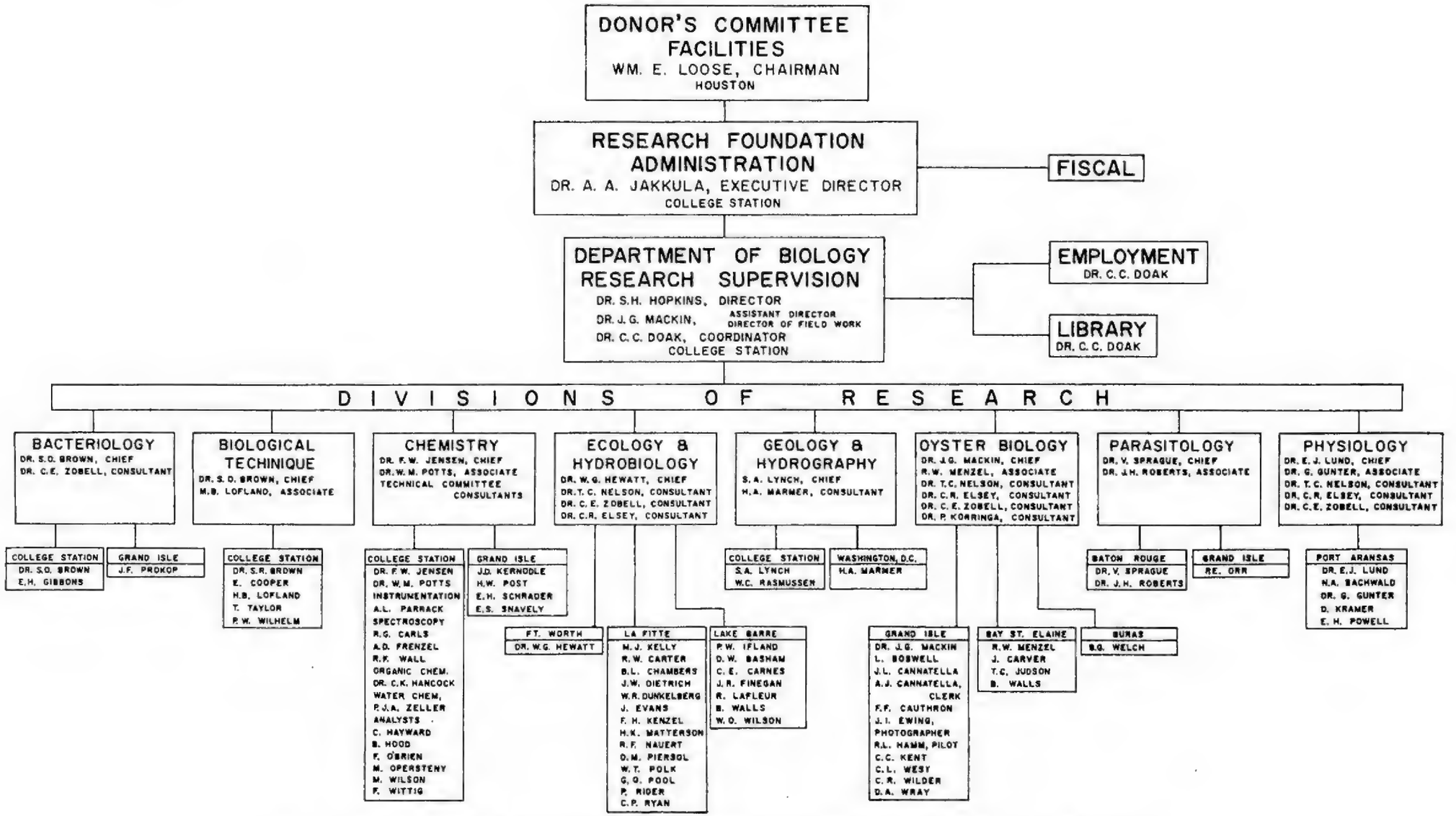

FIG. 1. Organization and personnel of Research Project 9 of the Texas A&M Research Foundation.

species of the fungus genus *Dermocystidium* Perez. This information was passed on to H. M. Owen and Albert Collier, who were independently studying the same disease. A description of the new species, *Dermocystidium marinum*, was published jointly by Mackin, Owen, and Collier in *Science* on March 31, 1950. Thus, after three years study, was accomplished the major objective of Project 9, the discovery of the cause of those mysterious oyster losses which were not due to any of the known lethal agencies such as storms, floods, or predators. But when Project 9 terminated on May 31, 1950, much of the epidemiology and pathology of the *Dermocystidium* disease remained unknown.

Project 23, sponsored by the six companies that sponsored Project 9 plus the Gulf Refining Company, started on June 1, 1950. The new Department of Oceanography (now Oceanography and Meteorology) at the A & M College of Texas was designated as the research agency. Project 23 was described as "maintenance of the Marine Laboratory at Grand Isle," but continuation of the oyster disease study was its main purpose. The nucleus of the staff consisted of four men, but more were employed for special work when necessary.

More than 400 reports and memoranda have resulted from the two oyster mortality projects. About 200 of these reported final results of researches; the rest were reports of trips, conferences, or the progress of uncompleted studies, and are of historical interest only.

JOHN G. MACKIN AND SEWELL H. HOPKINS

Studies on Oyster Mortality in Relation to Natural Environments and to Oil Fields in Louisiana[1]

J. G. MACKIN[2]

Department of Oceanography and Meteorology

AND

SEWELL H. HOPKINS

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[1] Oceanography and Meteorology Series from the Department of Oceanography and Meteorology, the A & M College of Texas. A report from Project 23E, Texas A & M Research Foundation, sponsored by Texaco Incorporated, Humble Oil and Refining Company, The California Company, Gulf Refining Company, Phillips Petroleum Company, and Shell Oil Company.

[2] Now Department of Biology, Agricultural and Mechanical College of Texas.

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Abstract

Field and laboratory studies and examination of historical records were made in the tidal waters of Louisiana to determine the possible role of operations of oil companies on mortality of commercial oysters, *Crassostrea virginica.* Periods of disastrously high mortality were found in past records, both before and after the beginning of oil operations in the area. According to official figures, 1940–1947 as a whole was a period of good production, but there was a decline in 1944–1947 due mainly to a sharp drop in St. Bernard Parish, outside the area of major oil operations.

Field studies showed that oysters had a consistently high mortality rate throughout the warmer half of the year in the study area, and that mortality rate increased with salinity increase within this area. There was no such correlation with proximity of oil fields.

Chemical studies revealed measurable quantities (0.001 to 0.010 percent dry weight) of unsaponifiable carbon tetrachloride extractives, called "hydrocarbons" by some chemists, in all mud samples tested, including many from locations where no pollution seemed possible. In some cases the quantity of such materials was correlated with the amount of plant debris in samples, and it was found that unsaponifiable material could be extracted by carbon tetrachloride directly from plants in areas distant from any source of petroleum. Within oil fields, the amounts of unsaponifiable carbon tetrachloride extractives were highest (up to 5.8 percent) in mud samples taken near bleedwater outlets and other known centers of pollution, and were correlated with high (up to 0.00578 percent) "pentane and heavier hydrocarbons" as measured independently by a commercial laboratory. Water samples showed minute quantities (approximately 1 ppm) of substances identifiable by routine analysis as "hydrocarbons" to be very widely distributed in Louisiana bay waters. These substances showed highest concentrations (up to 226 ppm) in oil field bleedwater and were still high (up to 7.6 ppm) in bay or bayou waters near the point of discharge, but decreased rapidly with distance from the source. "Hydrocarbon" content of mud and water was no higher in areas of high oyster mortality than in areas of low mortality. Calculations showed that bleedwater alone could not significantly increase the hydrocarbon content of mud or water on the commercial oyster beds where mortality occurred even if none were destroyed. Bacteriological studies showed that crude oil and its fractions were rapidly destroyed by bacteria living in Louisiana bay muds. Accidental oil spillages were found to cause enough oily taste to make nearby oysters unpalatable for periods ranging from several days to several weeks, but did not kill any oysters on commercial beds. In one case, however, a few intertidal oysters were killed by smothering.

Laboratory experiments showed that bleedwater, crude oil, water extracts or emulsions of crude oil, and the principal constituent of drilling mud (barium sulphate) had no effect on survival of oysters over periods of several months, when the concentrations tested were far above the maximum that could be maintained on an oyster bed. Quebracho, another constituent of drilling mud, caused some increase in mortality when used in concentrations much higher than could occur on an oyster bed. In physiological experiments, bleedwater and water extracts of crude oil slowed water pumping and filtering rates of oysters when used at concentrations much higher than could be expected in oyster-growing waters, but these effects were temporary and reversible.

In field experiments, spraying oysters repeatedly with crude oil and keeping them under a surface layer of crude oil did not affect survival or growth over a period of months. In other field experiments oysters were kept for months at various distances from bleedwater outlets, underwater natural gas discharges, and other sources of pollution. A discharge of 6600 barrels of bleedwater per day at Lake Barre increased mortality of oysters as far as 50 to 75 feet from the point of discharge, and apparently caused decrease of shell growth and glycogen storage in oysters as far as 150 feet away, but had no detectable effect at greater distances. Field experiments at Dog Lake, Caillou Island, Lake Pelto, and Bay Sainte Elaine oil fields did not show any adverse effect of bleedwater, natural gas, or small oil spillages upon the setting, survival, growth, or fattening of oysters. High explosives used for seismographic exploration in the manner prescribed by Louisiana law did not injure oysters either immediately or over a period of months.

It was concluded that oil production factors of the kinds tested could not be responsible for the oyster mortalities spread throughout the large area where damage was claimed. The widespread mortalities that did occur during the study period were correlated with high temperature and high salinity, but not with proximity to oil operations.

Introduction

Projects 9 and 23 of the Texas A & M Research Foundation were aimed at determining the cause or causes of mortality of oysters on the Gulf Coast, especially

in Louisiana and especially during the period beginning in 1940, when the oyster growers' claims of abnormal mortality began. This paper summarizes researches and findings on the Louisiana oyster industry and its history, the environmental conditions in the area where oyster mortality was claimed, and the interrelationships of the oil and oyster industries in that area. It is a summary, not an attempt to present complete data; it organizes, in a condensed form, data from a large number of separate reports that bear on one question: Has pollution by oil or oil well effluents, or seismographic exploration, caused mortality of oysters in Louisiana? For acknowledgment and an account of project administration (Fig. 1) see the preface to this volume.

Preliminary Studies and Definition of the Problem

The Research Foundation undertook (1) to determine whether or not mortalities of oysters in Louisiana were excessive, (2) to determine if mortality of oysters of considerable extent had occurred or was occurring, to determine the causes of the losses, and (3) if oil operations were responsible for any part of the mortalities, to determine what was to be done about it.

It was noted that recent accounts assumed that oyster mortalities were something new, and that the industry in Louisiana had previously been in a continuous state of good health. For this reason it was deemed wise to consult as many historical records as were available. The early records of the various state commissions having to do with oysters from the inception of state or parish supervision were found to be good sources of information. In a later section, data on oyster mortality from these historical sources are more fully detailed. Here suffice it to say that the record was puzzling in the extreme, for there appeared to be few periods within written history when the oyster industry was not in trouble. State production records show that there was no decline below the average in production at the time of alleged mortalities. The earliest records showed a steady decline in number of producing natural reefs, with here and there relatively catastrophic and rapid periods of decline during which production in whole areas was greatly reduced. Some of this loss occurred prior to 1900, but much was in the period from 1900 to 1920. In this period were witnessed several major attempts to rehabilitate an industry which considered itself in deep trouble at this early date. First the Gulf Biologic Station was established and devoted most of its efforts in its short life time (from 1902 to 1910) to detailed studies of the oyster industry and its troubles. In the same period the U. S. Bureau of Fisheries was sufficiently interested to send H. F. Moore (in 1898) and later Moore and T. E. B. Pope (in 1906–1908) to study means of developing a planting industry and to work out methods of rehabilitation of the moribund Barataria Bay industry.

As the century progressed a clearer picture of the situation began to take shape in published reports on the Louisiana oyster industry. First, it was apparent that depletion of natural reefs and the later troubles of the planted beds were associated primarily with high salinity of the oyster-producing waters. The occasional heavy losses to crevasses (Gunter, 1949) were well understood and these floods often did as much good as harm to the industry. But the unexplained losses were in nearly all cases associated with encroachment of Gulf waters carrying high salinities.

Second, the mortality waves were associated with high water temperature. Even when mortalities occurred in winter, they were associated with unseasonable warm weather.

Third, the mortality waves were selective of older oysters, and those used for counter stock were most affected. This is only another way of saying that mortality rates increase with age of the adult oyster. There was an apparent difference in death rates of seed oysters and those which had been on the bed for a year or more, the latter having a much higher mortality.

Fourth, the mortalities affecting oysters did not extend to the community of animals associated with them on the beds. Vigorously developing communities were reported on beds where 50 to 90 percent of the oysters died.

Certain data were compiled for the purpose of determining when and where mortalities occurred in the 1940 to 1947 period. These included (1) the locality of greatest alleged losses as indicated by the location of leases on which claims were filed and the amount of damages claimed, and (2) the period of greatest losses as indicated by the year for which claims were made. The data are contained in Tables 1 and 3.

A summary of the data which are presented in Table 1 shows that the damages were said to begin in 1940–41 with a quite modest total of less than $10,000 in claims.

TABLE 1

Claims for each of the years 1940 to 1948 in percent of the total $32,831,079 (1940–48)

Year	Percent of total
1940–41	0.03
1941–42	1.59
1942–43	2.18
1943–44	4.95
1944–45	5.69
1945–46	28.47
1946–47	55.53
1947–48	1.57

TABLE 2

Estimated value of total recorded oyster crop for Louisiana 1920–1949 at $5.00 per barrel*,†

1920—2,115,560	1930—3,230,770	1940—5,407,290
1921—2,204,160	1931—2,192,310	1941—4,889,035
1922—2,153,860	1932—2,157,680	1942—5,894,800
1923—2,666,655	1933—3,084,080	1943—5,389,325
1924—3,898,030	1934—4,716,040	1944—3,171,510
1925—3,365,645	1935—3,522,030	1945—3,142,145
1926—3,372,515	1936—3,838,485	1946—3,105,650
1927—3,791,115	1937—5,435,880	1947—3,306,295
1928—2,483,705	1938—2,880,250	1948—3,482,005
1929—3,858,170	1939—4,324,370	1949—4,133,700

* This estimate is admittedly much too high for most of the period of the table. It is used throughout to preserve uniformity.
† A Louisiana barrel is approximately 3.7 cubic feet and 0.106 cubic meters.

TABLE 3

Percent of claimed losses of oysters in different sectors of the Louisiana oyster-producing area
Total amount claimed was $32,831,079

St. Bernard Parish	Placquemines, E of Miss. R.	W. of Miss. R. & E of Grand Bayou	Greater Barataria Bay	Timbalier, Terrebonne Bay	W of Terrebonne Bay
Zero	3.0	16.0	67.0	14.0	Zero

NOTE: Because of the manner of filing the claims, the exact percentages belonging to each area could not be ascertained. The figures are approximate only.

In the year 1946–47 the claims reached their apex at over $18,000,000 or 55.5 percent of the total. It was clear that it was thought that there were catastrophic losses in the period 1945 to 1947 inclusive. A comparison of the dollar amounts with the estimated mean yearly value of the total Louisiana crop from 1920 to 1949 shows that the claims for the fiscal year 1945–46 totaled more than twice the mean, and the claims for 1946–47 were about four times the mean (Table 2). It was noted that according to Louisiana production statistics, the period from 1940 to 1947 did not produce less than the mean for all of the years, and the period from 1940 to 1943 had production significantly greater than the mean for the thirty years included in the table. Not all of the claims were for dead oysters. Damage to leases was also alleged but the comparative amounts of such losses were not generally stated.

The petitions pinpointed Barataria Bay, including its contiguous satellite bays and bayous, as the locale of the biggest part of the claimed losses. In Table 3 is a breakdown by areas, showing percentage of claimed losses in each. With 67 percent of the alleged losses in the Barataria basin, there was little trouble in deciding where to concentrate the efforts aimed at solution of the oyster mortality problem.

The views of oystermen were of importance in shaping the direction of researches. The Research Foundation made a study of the opinions of oystermen scattered throughout the area where heavy mortality was said to be a recurring annual event. In 1947, S. H. Hopkins interviewed 10 oystermen for opinions as to the causes of the mortalities, of whom four thought the operations of the oil companies were responsible for mortalities. Others made explanations which involved only natural causes.

Earlier Galtsoff (1942) had reported on a similar survey by Kavanaugh made in October, 1941. Kavanaugh talked to 13 oystermen, of whom two thought that pollution was one cause of the troubles. One of these named oil as the polluting agency. The other named both oil and sulphur bleedwater. Both named natural causes as responsible for part of the mortalities. Seven out of the 13 named natural causes; the others had no opinion on the losses. Of the natural causes given, high salinity was most often named. Conchs, bad water, "sick" seed, and lack of care in planting were also mentioned.

Chipman (1942) found only isolated cases of excessive mortality in the "high mortality" area of Louisiana in the summer of 1942. He concluded that one of these (in Bastian Bay) was caused by conches, but pointed out that conchs were not a problem in most oyster-growing sections. The other mortalities that he found were in Bayou Cholas and Bayou St. Denis.

A. E. Hopkins, representing the U. S. Fish and Wildlife Service, together with Louisiana officials, and representatives of the oyster and oil industries, made a survey of beds in the area from about Scofield Bay to Barataria Bay. This trip was in the period from February 11 to 15, 1947. Hopkins circulated a small memorandum of the findings. He found evidence of extensive losses of oysters. In his notes, he observed that the normal community of the oyster beds seemed to be unimpaired.

Certain facts of value emerged from the early interviews with oystermen. First, it was apparent that no one knew what was causing the alleged unusual mortalities. Second, it was evident that most of the trouble was in the Barataria Bay area, and third, that the greatest losses were believed to have occurred in the 1945–1947 period. Because of the great variety of opinion, it became necessary to study numerous individual sub-problems.

The Oyster Industry in Louisiana

A history of the oyster industry in Louisiana is a recital of trials and tribulations. The earliest researches (Moore, 1899; Zacherie, 1898; Cary, 1907a; Cage, 1904), and the latest (Owen, 1955; St. Amant, 1955; St. Amant *et al.*, 1956) were undertaken because of ills, imagined and actual, which beset the oyster industry.

The earliest troubles stemmed from a steadily increasing impoverishment of the natural reefs, on which the industry was based. Overfishing, according to early writers, was the prime cause of the failure of the natural reefs. It is believed, for reasons to be discussed later, that this was not the only reason for impoverishment of the reefs, but there can be little doubt that in some areas fishing pressure was intense.

In about 1886 (Moore, 1899) enterprising individuals began to make attempts at oyster culture, near the end of the Mississippi River delta. The seed came from Garden Island Bay, also at the end of the delta. Other plantings were made in the area of the "Salt Works" below Quarantine Bay, east of the Mississippi River.

As more bottom was made available to oystermen for planting purposes, the industry shifted its emphasis from tonging the natural reefs to extensive planting. Since the natural reefs became largely seed reefs under the new regime, the natural reef system never lost its importance, and natural reefs for the production of seed remain the backbone of the industry. But the plantings themselves developed troubles, basically the same as those which afflicted the seed reefs, with one difference. A planting cannot be "depleted," so depletion became recognized as mortality. The real planting period began about 1902 to 1904, when extensive holding of leased bottom was provided for by legislation. For a few years after 1904 the oystermen were allowed to use dredges for taking seed from state-operated reefs, and the general picture was one of a developing, vigorous industry based on seed planting and continued use of the natural reefs. The following sections give a more detailed account of the production based on the natural reefs and on the later seed and shell planting. Information is based on published accounts of researches and the semi-annual reports of the various state agencies having to do with the oyster bottoms.

The Natural Reefs

H. F. Moore (1899) made the first survey of the natural reefs of Louisiana, but failed to estimate the acreage involved. He found extensive natural reefs, as well as many extinct reefs, all the way from the Louisiana Marsh to Atchafalaya Bay. Moore suggested that a natural reef should be defined as follows: "A natural oyster reef, bar, or bed is an area of not less than 500 square yards of the bottom of any body of water upon which oysters are found or have been found within a term of five years—in quantities which would warrant taking them for profit by means of tongs." The dimensions of such a natural reef might be 50 by 10 yards and the area only about 1/10 acre. Moore's definition anticipated the time when leasing on a large scale would begin, and decisions would have to be made as to whether a section of bottom was available for leasing.

Payne (1912) made a hurried survey of the natural reefs in the fall of 1910. He found a total of 62,740 acres which presumably came within Moore's definition. Most of this (53,200 acres) was in Sister Lake, Bay Junop, and the Point Au Fer reef in

Atchafalaya Bay, which probably included Marsh Island reef. This left a small amount for the Louisiana Marsh (3,040 acres), and for California and Quarantine Bays (6,000 acres), and left none for Barataria Bay, Timbalier Bay, and Terrebonne Bay, the area west of Terrebonne Bay to Sister Lake, or any satellite bays or bayous. It seems clear that Payne ignored vast areas of low grade coon oysters and all areas of sparse growth. Since he was mainly interested in developing a planting industry, it served his purpose to reserve these areas for private leasing. Cary (1907b), just four years prior to Payne's survey, had studied all of the natural reefs from Sister Lake to Raccourci, including Terrebonne Bay and contiguous waters, and had found extensive natural reefs. Also, McConnell (1932) stated that there were 50,000 acres of natural reefs east of the Mississippi River in St. Bernard and Placquemines Parishes. While this may be an overstatement, it is probably a much better estimate than was Payne's "survey" of 9000 acres.

There has never been a real survey of Louisiana's natural oyster reefs and no one will ever know how extensive was the resource prior to the construction of the Mississippi levees. Before the turn of the century (Moore, 1899), bays like Barataria and Terrebonne reportedly contained only relict reefs. The estimates of Payne and McConnell were made after Gates (1910) had declared that natural reefs were a thing of the past. This certainly was not true either, as is proved by the present phenomenal production of the Placquemines seed reefs in California, Quarantine, Black, and other bays. But the lack of agreement shows that no one ever really knew the magnitude of the resource.

Locations of the Natural Reefs

All of the Louisiana saline marshes support natural growths of oysters. Most of these are patches of coon oysters, some small, some large. In high salinity areas these patches are apt to be intertidal, and are almost always in the bayous and small bays. Many are in marsh ponds only narrowly connected by tidal runs to the larger bays. In waters of lower salinity the natural oyster beds are subtidal, and may be in the bottom of deep scoured bayous. Most often these small oyster beds are developed on the old *Rangia* beds; such a bed may produce both oysters and clams in scanty numbers.

Generally, by the "natural" reefs, authors mean those of large extent, in large bodies of water. At present these exist in the following areas:

(1) In Mississippi Sound north of the Louisiana Marsh and in the Louisiana Marsh proper there are still many extensive natural reefs. Most of these at present have only thinly scattered oysters and many do not qualify for the name of reef, since there is no solid shell substrate.

(2) In the area from Mozambique Point to California Point, east of the Mississippi River in California, American, Quarantine and Black Bay, and in some others there are actual hard reefs. They are highly productive, but have not always been so. They are dredged for seed annually, and many are almost literally left stripped. Culling is not practiced and the reefs take the hardest abuse imaginable. In spite of this rough treatment they rebound in a very short time. At present they provide most of the seed used by Louisiana planters.

(3) At the end of the Mississippi River delta there are several small bays with very low salinity into which the Mississippi pours huge quantities of water in flood periods. Under the predominantly fresh surface water, tongues of salty Gulf water push in along

the bottom and maintain small hard reefs in these exposed bays. These are economically insignificant, but of great biological interest.

(4) Westward from the Mississippi as far west as Dog Lake and Hackberry Bay there are innumerable small natural reefs. Most of these are individually unimportant but in the aggregate they produce many fine oysters. Nearly all are in the bayous and small bays to the north of the main water bodies. They can exist only in those water bodies that are periodically flushed by local freshets. This flushing reduces both conchs and fungus disease to low levels and thus increases longevity of the oysters so that they reach market maturity in satisfactory quantities. Much planting is done in the same small water bodies, using the local seed.

(5) In Sister Lake, Bay Junop, and the neighboring bays there were once major natural reefs. These have been nursed along by the Division of Water Bottoms and still contribute respectable quantities of the seed oysters used in areas too far west to depend on the reefs east of the Mississippi. In late years (St. Amant, 1955, St. Amant *et al.*, 1956) Sister Lake, because of increasing salinity, has largely broken down as a natural producing area.

(6) In Atchafalaya Bay, the Point Au Fer reef, the largest in Louisiana, is continuous on the far west with the Marsh Island reefs. A large part of the Point Au Fer reef is now dead because of increasing flow of fresh water from the Atchafalaya River. For some years it has been mined for shell. The western end still has live oysters and the Marsh Island reef has produced seed of an inferior sort for many years. Most of the Atchafalaya reefs are massive, solid, and stand high above the surrounding bottom. They are useless for oyster production but are valuable as a source of basic information.

(7) In Vermilion Bay and others to the west of Atchafalaya Bay, there are intermittent reefs all the way to the Sabine River. The Vermilion Bay reefs in recent years have shown signs of developing into an economic asset but do not compare with the producing areas farther east.

Depletion of the Natural Reefs

Moore (1899) was the first to publish data on the decline of production of the natural reefs of Louisiana. His mission in Louisiana was evidently to develop sufficient knowledge of quantities of oysters available, oystering methods, and basic ills to advise the Legislature in framing oyster law. The emphasis placed by Moore on the declining natural reef production and the desirability of developing a healthy planting industry shows that his chief concern was the apparent failure of the natural reefs to meet the needs of an expanding industry.

Cage (1904) described the oyster industry of the period prior to 1870. According to him, depletion of the natural reefs was far advanced prior to that date. He stated that the reefs in Placquemines Parish were "exterminated" first, which led the oystermen from there to invade the more westerly Parishes where the reefs were "little used." They apparently used the reefs in Barataria, Timbalier, and Terrebonne bays for obtaining seed, planting the oysters in the Placquemines beds. Barataria reefs followed the Placquemines route to depletion, and in turn those in Timbalier Bay and Terrebonne Bay.

Dymond (1904) stated that he knew of "hundreds of natural reefs that have been completely wiped out," but he failed to point out the locations. It is assumed that he had

in mind the Placquemines and Jefferson Parish reefs. In 1906 the oyster commission of Louisiana reported that natural (seed) reefs east of the Mississippi in Placquemines Parish had suffered because of extensions of the levee system on that side. It was also stated that because of lack of food, the oysters had for two years been very poor. This is probably the first report that coupled increased salinity, resulting from the levee system, with depletion of oyster reefs. Baylor (1904) had advocated extensions of the levee system as a means of protecting oysters from destruction by fresh water. By way of contrast, Breaux *et al.* (1908) in the oyster commission report for 1906 to 1908 stated that "It cannot be said that the natural oyster reefs of this state are in danger of depletion." Cary (1907b) made an extensive study of oyster reefs in Terrebonne Parish, from Sister Lake eastward to Lake Raccourci. He listed 32 bays and bayous in which he found depleted natural reefs; many of these had no oysters on them at all. Moore (1899) had briefly surveyed this same region, and Cary pointed out several dead or dying reefs which Moore had named as good producers eight years earlier. Gates (1910) stated that nearly all natural reefs in Louisiana were then depleted, which was obviously an overstatement. Gates was in favor of leasing all natural reefs, even if they were producing, which probably explains his exaggeration. It was in 1910 that Payne (1912) surveyed the natural reefs and came up with his figure of 62,740 acres. Payne (1914) reported that the natural reefs of St. Bernard Parish (the Louisiana Marsh) were depleted and that some of these had been leased for planting purposes.

A most important loss to the industry was reported by Payne (1914). In 1904 Bayou Lafourche, an important distributary of the Mississippi, was severed from the river. Formerly, it fed fresh water to the important seed reefs of the northeast part of Timbalier Bay. As a result of increasing salinity following closure of the bayou, these reefs deteriorated badly. They are not dead at the present date, but production is marginal and unpredictable.

There are subsequent references to decline in production of the natural reefs of Louisiana, but most of them do not distinguish between planted beds and natural reefs. Statements of early authors showed clearly that a serious decline had occurred over large areas, and just as clearly that the natural growth of oysters was prolific in many other areas. As reefs disappeared, usually in high salinity areas, new ones appeared in lower salinity areas and, as Payne (1912) remarked, the industry appeared to be moving inland.

Causes of Depletion of the Natural Reefs

Moore (1899) believed that the destruction of the natural reefs was due to overfishing coupled with failure to cull out and return shells and young oysters to the reefs. Cage (1904) thought that failure to observe spawning seasons and failure to cull were responsible for destruction of the reefs. Cary (1906, 1907a) thought that local freshets, boring sponges, conchs, and overfishing were causes of depletion. Moore and Pope (1910) made experimental plantings which were successful in low salinity waters but uniformly failures in high salinity areas. Payne (1912) blamed destruction of reefs in Caminada and Timbalier Bays on closure of Bayou Lafourche, and overfishing for depletion of reefs in Bay Junop and Sister Lake. He later (1920) attributed the decline of reefs in Placquemines east of the Mississippi to high salinity, and the destruction of Barataria reefs to the same factor.

It is difficult to agree that overfishing was the major cause of depletion in the early years of the industry. First, only tongs were used for taking oysters, and it is not believed to be commercially feasible to tong a reef to the point of biological extinction. The production recorded before the turn of the century was not great enough to make serious inroads on the natural supply. If Payne was right in his survey, and there were 12,720 acres of natural reef (not counting Point au Fer), a rough estimate of fishing pressure can be made. According to Gunter (1949) the average annual production of oysters between 1880 and 1900 was about 236,000 barrels. The acreage of the natural reefs must have been much greater in 1880 than Payne's 1910 survey showed, and it is estimated that it was probably twice as great. If that is true there were about 25,000 acres of natural reef to begin with, which were being worked at an annual rate of 236,000 barrels in the 20-year period from 1880 to 1900. Each acre therefore produced less than 10 barrels annually. This is not a great fishing pressure, especially in view of the fact that even prior to 1900 some of the production was from shell plantings.

On the other hand, the observed correlations of high salinity and depletion appear to present at least a workable hypothesis. High salinity in itself is not detrimental to oysters, as numerous studies have shown. Oysters in Louisiana grow faster, spawn more prolifically, and set in larger numbers in high salinity waters, up to oceanic salinity concentrations, than they do in low salinities; but competition, predation, and disease impose a high annual death rate on both immature and mature oysters in highly saline waters. Rises in salinity paralleled the construction of more efficient and more extensive levees along the lower Mississippi River (Gunter, 1952). The higher and more stable salinity made the oyster beds more favorable habitats for other organisms, including competitors, predators, parasites, and other enemies, and therefore less favorable for oysters. Hence the increase in oyster mortality accompanying the increase in salinity.

The Development of the Planting Industry

The beginning of the planting system in Louisiana occurred in the period between the end of the Civil War and 1885. Oystermen bedded oysters, which they took from the natural reefs, on limited areas of hard bottom near their camps. They had no lease or title to these "bedding grounds." Bedding of oysters did not mean planting for growth or cultivation in any modern sense. Oysters tonged from the natural reefs were piled on a limited area of clean shell bottom, so that they could be quickly reloaded to take to market when a boatload was accumulated.

It is not certain just when this practice developed into a true planting system. Moore (1899) gives credit for pioneering in oyster cultivation to Louis Esponger, who planted seed in Whale Bay in 1885. The seed for this planting came from Garden Island Bay. Both bays were at the extreme end of the Mississippi Delta, and both are now silted into deltaic plain. Esponger's effort may have been preceded by efforts of others who planted in the area called the Salt Works, just to the south and east of Quarantine Bay on the east of the Mississippi. At one time, these plantings, added to production from natural reefs in the Louisiana Marsh, supplied most of the New Orleans trade. It appears certain that planting first developed in Placquemines Parish, where the natural reefs were first depleted.

The Louisiana Legislature first made bottom available for lease in 1886, but the amount was only three acres per person. It seems that this amount was made available

only to allow oystermen to control their holding or "bedding" grounds to prevent theft of their oysters while they completed their loads. In 1892, the amount available for lease was increased to ten acres. By leasing in the name of relatives oystermen could accumulate sufficient acreage of bottom to permit actual planting, and it is believed that the first real impetus to planting shell as well as seed developed in the period from 1892 to 1902. In this latter year the allowable holding per person was increased to 20 acres and in 1904 the limit was extended to permit one individual or corporation to hold 1000 acres. This is the limit today.

According to Moore (1899) the first "plantings" were made on areas as small as 50 feet square and oysters were piled as much as a foot deep on the bottom. This obviously was not planting but a step in marketing. It is interesting that this is still a method in wide use in Louisiana, and is sometimes costly to the oysterman.

Moore also described planting of culch in Placquemines Parish. Shell or other culch was planted in shallow water from a few inches to 18 inches maximum depth and along the shores of bays and bayous. Louisiana bays and bayous are usually very soft in the middle, while the margins are of stiff mud supported by heavy concentrations of partially decayed plant materials derived from the marsh. Young oysters caught on culch along the margins were culled out at the end of the year and planted in deeper water as singles. As seed they were crowded and of typical coon oyster type, but after culling they quickly developed good shape and size.

Louisiana oysters at that time, as at present, grew well in salinities lower than the lowest in which oysters could survive in more northern climes (Moore, 1899; Mackin, 1956a). This is in agreement with the theory that marine organisms extend their ranges into lower salinity in warm countries (Hesse, Allee and Schmidt, 1937). But even at that time most oyster leases were in high salinity areas, as evidenced by the trouble with conchs.

Moore (1899) estimated that in 1898 there were 500 oystermen operating in the area between Bay Jacque and Bastian Bay. Most of these planted oysters, for Moore remarked that the natural beds of Placquemines Parish west of the river were "limited in extent and of but little importance." Oystermen planted 600 to 800 sacks of seed oysters per acre of bottom and, according to Moore, sometimes harvested two sacks for one at the end of a year. Two years from setting time sufficed to mature oysters for market.

A lesser planting area was developed in northeast Timbalier Bay, using the seed growing in the same area. There was no estimate of number of men engaged in this industry. Farther west in Terrebonne Bay and westward to the Atchafalaya about 32 oystermen were engaged in planting, a relatively small number for this very large area.

Dymond (1904) summed up the planting industry from its beginning to 1902. In all the period prior to that time 512 leases had been taken out by oystermen, totaling 2820 acres. The average size of these leases was 5.5 acres, suggesting that most oysters "planted" were only held for marketing. In 1904 a rapid expansion began when the leasing of 1000 acres was permitted and the number of leases increased to 1194 with an average size of 19.5 acres. The total acreage increased in two years from 2820 to 21,853. Although the leased area increased nearly 700 percent, the production from 1903 (reflecting 1902 planting) to 1905 (reflecting 1904 planting) increased only 42 percent. This was because most production still came from natural reefs. As late as 1908 the oyster commission reported that more than half of all production was still

from natural state-owned reefs. In 1907 total production was 933,960 barrels. If we assume that about 40 percent of this was from plantings, 374,000 barrels were produced from about 22,000 acres of bottom which is an average per acre of 17 barrels.

Leasing for subsequent years varied from about 13,000 acres in 1935 to 48,000 acres in 1957. During the period of the multi-million dollar suits against the oil companies from 1940 to 1947 the leased acreage for oyster planting rose steadily from 14,749 to 23,279. The fluctuations in leased acreage for Louisiana are contained in the graph, Fig. 2. After the planting system was established the year of smallest production was in 1920 (see Fig. 3), in which year there were under lease about 20,000 acres of bottom. The year of the greatest production was 1942 and in this year the acreage under lease was only about 16,000 acres. The recorded figures show no close relation between acreage under lease and total production.

In the years from the end of the depression in about 1939 to the present, acreage under lease has generally increased. In the last year here recorded (1957) there were about 48,000 acres under lease. As far back as 1912 the Board of Commissioners for the Protection of Birds, Game and Fish (see Miller, Grace and Dodson, 1912) estimated that there were about 472,000 acres of good bottom in Louisiana. If this is a correct estimate Louisiana planters have never utilized more than about 10 percent of the available good bottom. Our own experience indicates that 472,000 acres was a gross overestimate. Many oystermen in Louisiana plant on bottom too soft or otherwise unfit for oyster culture. This was true as far back as 1898 (Moore, 1899). The writers believe that much of the bottom used in the early part of the planting era is now worthless, because of deterioration of the bottom or increased salinity of the water, and that oystermen have shifted most of their planting to new bottoms formed by the natural disintegration of shorelines.

As the planting industry developed and production from natural reefs decreased two trends became apparent. One was the trend toward planting more seed and less shell. Louisiana oystermen in the early days of planting used both the seed planting and the shell planting methods. Although the shell planting method was said to be highly successful in the early part of the century, it nevertheless was largely abandoned in later years and a decided preference was given to seed planting. Shell planting by oystermen is still done but is now a relatively unimportant part of the industry, and has been so for many years except the plantings of shell in the Louisiana Marsh by the canning industry. Emphasis on seed planting has always thrown a heavy strain on the seed reefs and has required an increasing amount of shell planting for "rehabilitation" purposes. Originally rehabilitation was a correct word to use. More recently the State has taken over the function of planting shells on barren bottom. These shells are not for either maintenance or development of permanent seed reefs. When oysters attached to the culch are nearly large enough for market, they may be dredged by the oystermen from the State "seed" reservations, transplanted to the oystermen's leases, and shortly thereafter dredged for market. Thus much of the responsibility for success in oyster culture is now the State's, and the cost of all failures falls to the State.

The other trend has been toward short-term planting methods. As early as 1920 (Payne, 1920), seed oysters planted in Barataria Bay were kept on the beds for as short a period as three to four months. McConnell (1932) also mentioned that oysters were kept on the beds only six months as early as 1932. This applied to large seed, small seed being left for a year. The short period usually was designed to include only cool months,

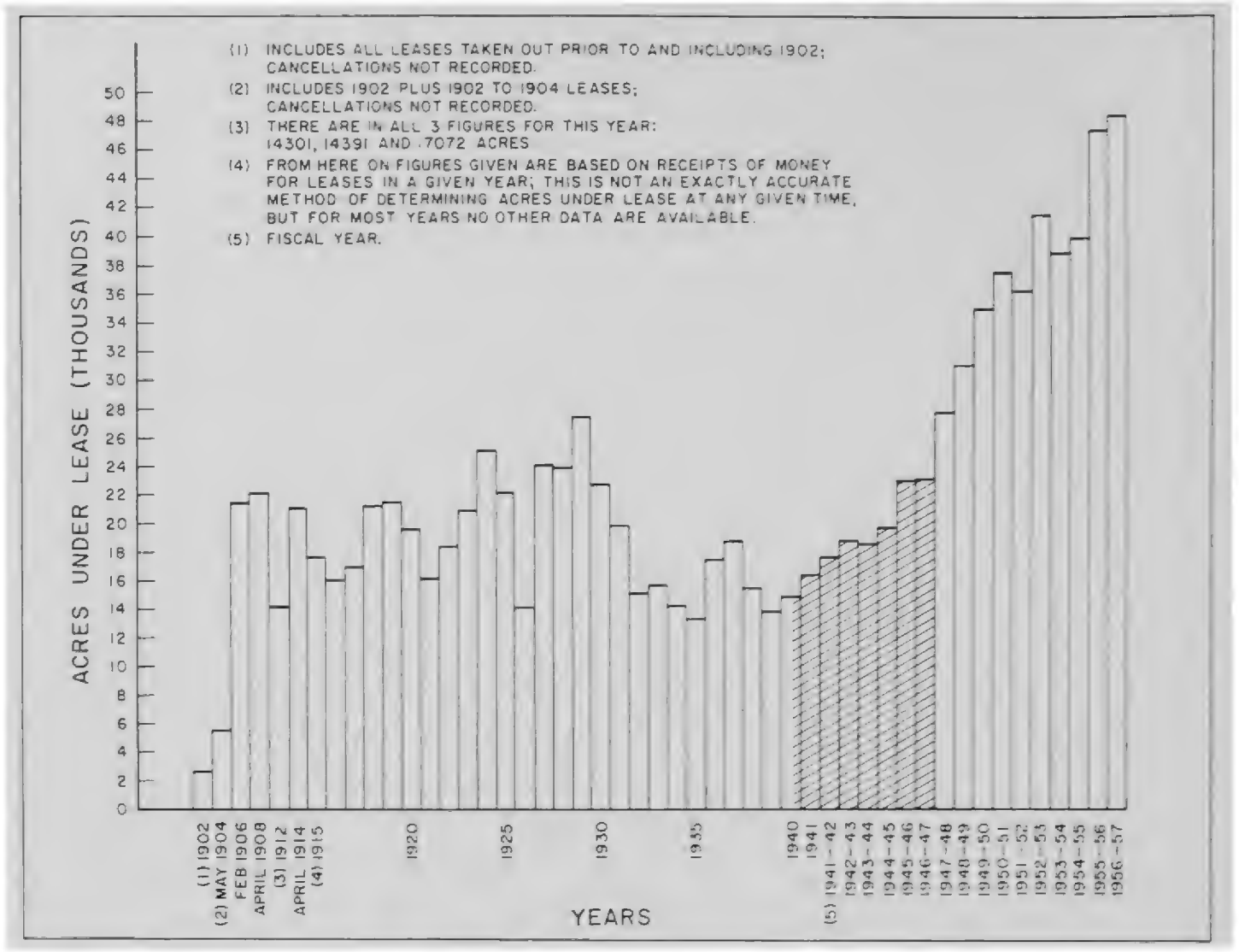

FIG. 2. Fluctuations in acreage leased for oyster planting in Louisiana, 1902–1957. Shaded area includes years in which pollution damage was claimed.

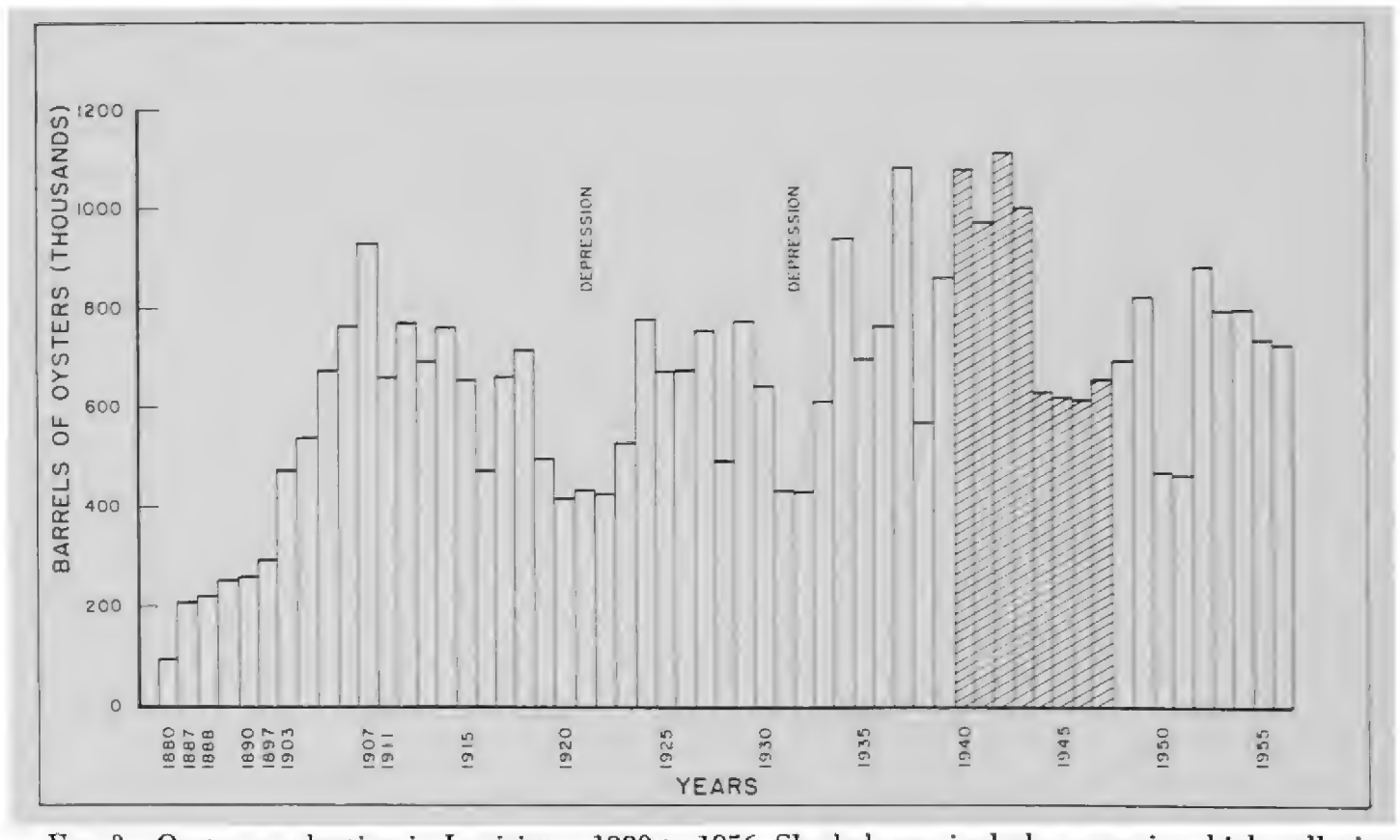

FIG. 3. Oyster production in Louisiana, 1880 to 1956. Shaded area includes years in which pollution damage was claimed.

since the growth and fattening of Louisiana oysters is best in winter and the high mortality of summer periods is avoided. The short planting season required high salinity

for rapid shell growth, and a large seed oyster which was nearing maturity. Under these conditions planting for periods of less than three months produced a fair grade of raw shucked oyster, some rather small counter oysters, and quantities of very fine canning stock. As mortality rates in counter stock increased with the years, greater and greater emphasis was placed on the short-term planting period.

From the beginning of the planting industry, in spite of the more sensational losses from floods, most reports have connected decreased production with increasing salinity in Louisiana bays, the same common denominator which underlay the disintegration of the natural reef industry. Cary (1907b) discussed mortalities of oysters on planted beds in Terrebonne Parish. Mortalities were great enough in Bay Wilson that oystermen were abandoning leased ground. Cary thought that Bay Wilson oysters were destroyed by the prolific growth of the boring sponge, a certain sign of high salinities. Payne (1914) attributed the collapse of the industry in northeast Timbalier Bay and Caminada Bay to high salinity caused by closure of Bayou Lafourche to Mississippi water in 1904. Reports of the Conservation Commission returned repeatedly to the theme that high salinity and high mortality were correlated (Alexander, 1922; Payne, 1920). The movement inland of the planting industry was a result of encroaching high-salinity Gulf water and disintegration of shorelines. In 1932 McConnell attributed extensive losses of counter oysters to a combination of economic depression (causing delay in marketing) and warm weather during winter. Extensive losses of oysters in Terrebonne Bay in 1931–32 and 1932–33 were connected by Prytherch (1933) to warm weather in winter and to boring clam and sponge. Some of the mortalities were said to have occurred while oysters were in transit to market, after being held on holding grounds. McConnell (1934), apparently referring to the same mortality, set the loss at 300,000 barrels. In the same year (1932) 250,000 barrels were lost in the upper Barataria region. This loss was preceded and followed by extensive drought, with a single very heavy rain about at the time of loss. Gunter (1952) reviewed the literature on mortalities of Louisiana oysters including those said to be correlated with rising salinities. Owen (1953) found that in the period from 1932 to 1950, production of oysters dropped significantly following periods of high temperature and low rainfall.

Not less significant was the accumulating evidence that mortalities were also associated with high-temperature months, and that mature oysters had greater mortalities than did young oysters. As time went on it became more and more difficult to produce counter oysters, i.e., the largest (and therefore oldest) oysters. The fact that mortalities on planted beds were selective of counter oysters was mentioned by McConnell (1934) for the period 1932–33, when all Louisiana was affected by warm weather mortalities and poor condition. Both 1932 and 1933 were dry years; 15 out of the 24 months had deficits in rainfall (Hewatt, 1955). Galtsoff *et al.* (1935) also called attention to the disproportionate loss of older oysters where young oysters were unaffected. McConnell (1940, 1942) returned to this thesis and pointed to losses of counter oysters in 1939, 1940, and 1941 which were correlated with high-temperature months as well as high salinity. He stated that there was an increasing annual loss of large oysters with little loss of young oysters.

A. E. Hopkins (1947) concluded that there were heavy summer mortalities of oysters in the bays of the west flank of the Mississippi, including Barataria Bay, which probably started as far back as 1939. Hopkins observed, in this survey, that animals of the oyster community other than the oysters themselves did not suffer unusual mortalities.

Production Statistics

Gunter (1949) presented a table containing statistics of production of oysters in Louisiana from 1897 to 1947. Seferovich (1938) reported on some early years (1880 to 1890). His figures were from U. S. Bureau of Fisheries reports and were converted to barrels. Figures for the period from 1948 to 1955 have been taken from reports of the Louisiana Department of Wildlife and Fisheries. From these sources the graph, Fig. 3, was made. The data are also shown in Table 4.

Table 4

Records of production of oysters in Louisiana. Records for years in the 19th Century are from Seferovich (1938), and are approximate only. Records for 1956 are from Gulf Fisheries Leaflet, 1956. All others are from data published by the State of Louisiana through various agencies. Years are fiscal years ending in the year indicated.

Year	Barrels*	Year	Barrels	Year	Barrels
1880	92,000	1919	503,947	1938	576,050
1887	210,000	1920	423,112	1939	864,874
1888	220,000	1921	440,852	1940	1,081,458
1889	257,000	1922	430,772	1941	977,807
1890	260,000	1923	533,331	1942	1,178,960
1897	295,135	1924	779,606	1943	1,077,865
1903	472,191	1925	673,129	1944	634,302
1904	540,192	1926	674,503	1945	628,429
1905	673,060	1927	758,223	1946	621,130
1906	765,002	1928	496,741	1947	661,259
1907	933,960	1929	771,634	1948	696,401
1911	661,094	1930	646,154	1949	826,740
1912	771,111	1931	438,462	1950	470,600
1913	693,689	1932	431,536	1951	467,074
1914	762,880	1933	616,816	1952	885,716
1915	658,255	1934	943,208	1953	793,074
1916	479,462	1935	704,406	1954	797,224
1917	632,785	1936	767,697	1955	739,516
1918	710,068	1937	1,087,176	1956	728,038

* The Louisiana barrel is approximately 3.7 cubic feet or 0.106 cubic meters.

Used with discretion and a thorough background knowledge of Louisiana industry these figures are of some value. The total oyster production figures show that Louisiana has through the years maintained a high level of production, in spite of the fact that in every decade of this century state officials have been pointing with alarm.

Production in Jefferson Parish Compared with Production in the Louisiana Marsh (St. Bernard Parish)

In this study one primary objective was to determine the validity of the claims that oil production was a cause of mortality, and thereby of decreased production of oysters. If the claims in the 1940 to 1947 period were valid, Barataria Bay (Jefferson Parish) should show a steep decline in production in that period, which would not be true of areas where oil operations were not developed and where no claims for damage were made. Such an area is in the Louisiana Marsh (St. Bernard Parish). If parallel decline occurred in St. Bernard Parish, the fact would indicate that factors other than oil production were effective. The production statistics of Jefferson and St. Bernard Parishes were studied to determine what were the trends, and whether or not fluctuations in production were similar in the two parishes.

As a basis for comparison, it was assumed that the production during the decade of 1930 to 1940 was representative. Comparative data are available for eight of those years. From 1932 to 1939, St. Bernard Parish averaged 533,192 barrels of oysters annually and Jefferson averaged 60,874 barrels. These figures were assumed to represent 100 percent production for each of the two parishes, and production for the years 1940 to 1953 were plotted as percentages of these figures. The data are presented in Fig. 4. The similarity of the two graphs is obvious. Both parishes produced above normal in the first years of the decade from 1940 to 1943, both declined steeply in 1944 and 1945, and both remained at a low level to 1951. Very remarkably, both regained normalcy in 1952, and declined again in 1953. The official oyster production figures therefore indicate that the heavy mortalities were not confined to areas of oil production, but affected non-oil-producing territory also.

The actual loss in St. Bernard Parish was more than eight times as great as it was in Jefferson Parish. Percentage loss was about the same, but the production plunge for the state as a whole, in the 1944 to 1950 period, was largely due to an enormous loss in St. Bernard Parish where at that time the oil industry was in its infancy. There were no oil wells in the oyster-producing area of St. Bernard Parish.

In the last decade, several spot checks of production in Jefferson Parish have been made. These indicate that the quantity of oysters actually produced in this parish in recent years is much larger than that indicated by official state figures for the same years. For example, the senior author (Mackin, 1951a) checked one planted bed in

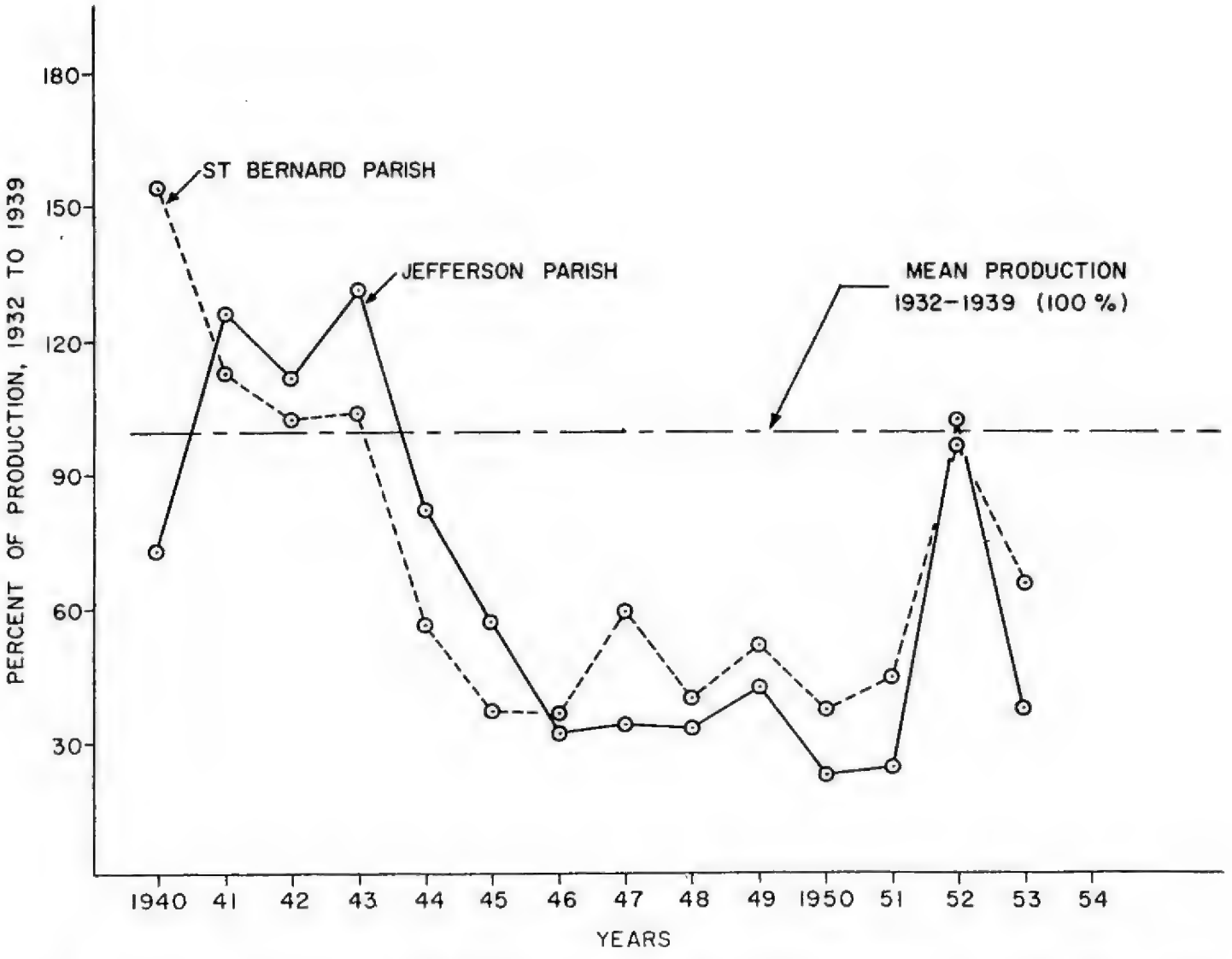

FIG. 4. Oyster production in St. Bernard Parish as compared with Jefferson Parish, 1940 to 1953.

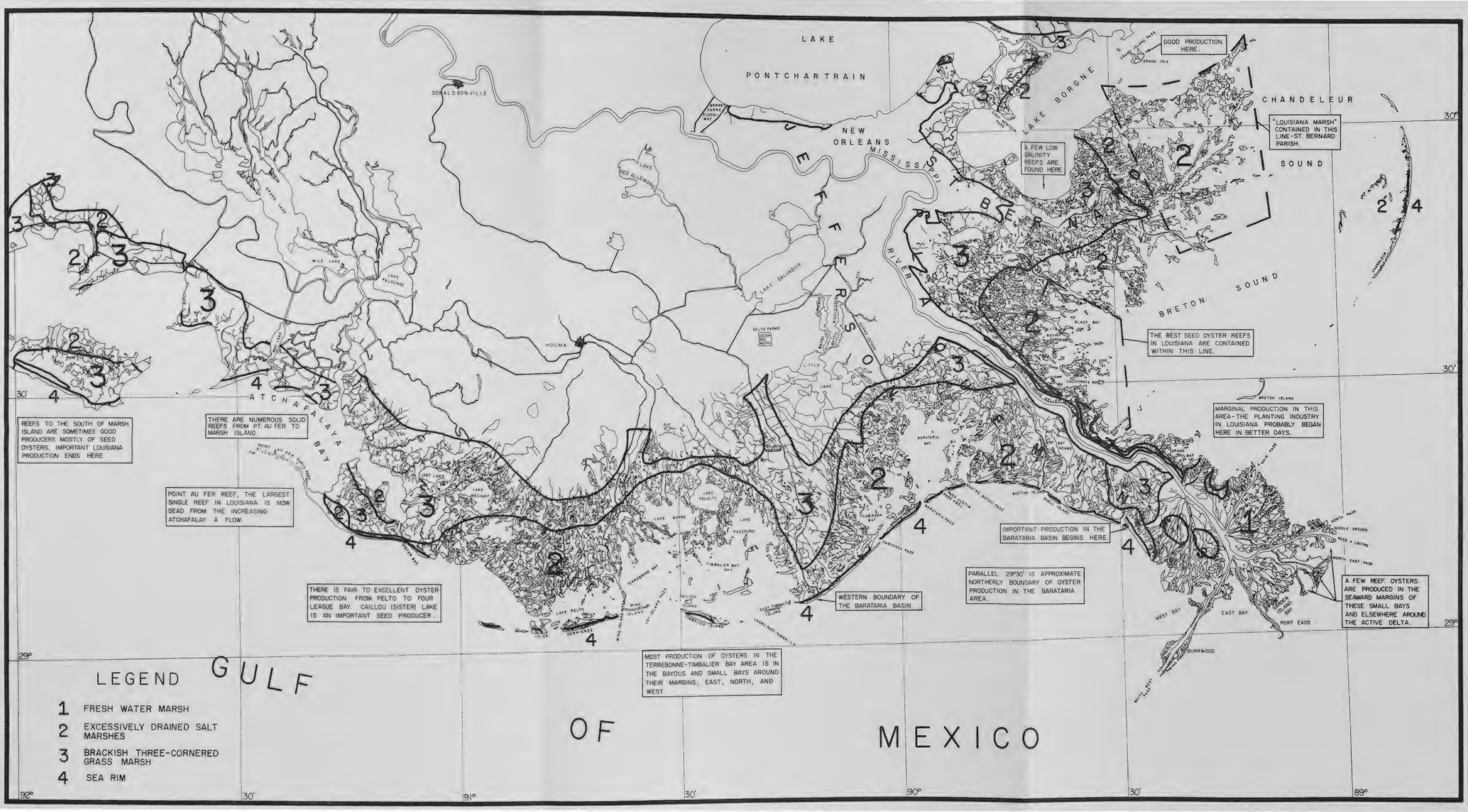
LAKE
PONTCHARTRAIN
DONALDSONVILLE
NEW
ORLEANS
MISSISSIPPI
RIVER
LAKE BORGNE
CHANDELEUR
SOUND
BRETON
SOUND
LAKE
DES ALLEMANDS
LAKE SALVADOR
HOUMA
DELTA FARMS
ATCHAFALAYA
BAY
TERREBONNE
BAY
TIMBALIER BAY
LAKE PELTO
ISLES
DERNIERES
GULF
OF
MEXICO
GOOD PRODUCTION HERE.
A FEW LOW SALINITY REEFS ARE FOUND HERE
"LOUISIANA MARSH" CONTAINED IN THIS LINE-ST. BERNARD PARISH.
THE BEST SEED OYSTER REEFS IN LOUISIANA ARE CONTAINED WITHIN THIS LINE.
MARGINAL PRODUCTION IN THIS AREA-THE PLANTING INDUSTRY IN LOUISIANA PROBABLY BEGAN HERE IN BETTER DAYS.
REEFS TO THE SOUTH OF MARSH ISLAND ARE SOMETIMES GOOD PRODUCERS MOSTLY OF SEED OYSTERS. IMPORTANT LOUISIANA PRODUCTION ENDS HERE
THERE ARE NUMEROUS SOLID REEFS FROM PT. AU FER TO MARSH ISLAND.
POINT AU FER REEF, THE LARGEST SINGLE REEF IN LOUISIANA IS NOW DEAD FROM THE INCREASING ATCHAFALAY A FLOW.
IMPORTANT PRODUCTION IN THE BARATARIA BASIN BEGINS HERE.
PARALLEL 29°30' IS APPROXIMATE NORTHERLY BOUNDARY OF OYSTER PRODUCTION IN THE BARATARIA AREA.
A FEW REEF OYSTERS ARE PRODUCED IN THE SEAWARD MARGINS OF THESE SMALL BAYS AND ELSEWHERE AROUND THE ACTIVE DELTA.
THERE IS FAIR TO EXCELLENT OYSTER PRODUCTION FROM PELTO TO FOUR LEAGUE BAY. CAILLOU (SISTER) LAKE IS AN IMPORTANT SEED PRODUCER.
WESTERN BOUNDARY OF THE BARATARIA BASIN.
MOST PRODUCTION OF OYSTERS IN THE TERREBONNE-TIMBALIER BAY AREA IS IN THE BAYOUS AND SMALL BAYS AROUND THEIR MARGINS; EAST, NORTH, AND WEST.
LEGEND
1 FRESH WATER MARSH
2 EXCESSIVELY DRAINED SALT MARSHES
3 BRACKISH THREE-CORNERED GRASS MARSH
4 SEA RIM
30°
29°
92°
91°
90°
89°

FIG. 5. Map showing principal oyster-producing areas of Louisiana, and ecological areas as shown by O'Neil (1949). Oyster-producing areas are about co-extensive with O'Neil's area 2, the excessively drained salt-marshes.

TABLE 5

Summary of shell and seed plantings for production of seed oysters in Louisiana

Year	Location	Amount planted barrels
May 1917	Sister Lake	16,238
Aug. 1917	Cabbage Reef	5,975
1917 (?)	Not stated	137,114
July 1919	Not stated	1,200
May 1929	Mississippi Sound; Turkey Bayou	60,000
May, June 1930	Bay Boudreaux; Fox Bay	45,000
Spring 1931	Fox Bay	35,000
1932 (?)	Quarantine	15,000
1933 (?)	Bay Boudreaux	55,000
Spring 1934	Nigger Point	75,000
1935	Quarantine Bay	20,000
1936–37	Not stated	150,000
1938	Mississippi Sound; Half Moon Island	34,020
Spring 1939	Mississippi Sound; Half Moon Island	58,711
Summer 1941	Sister Lake	50,000
Spring 1941	Mississippi Sound; Half Moon Island	70,000
Summer 1943	West of Mississippi R.; Sister Lake	91,632
1943 (?)	East of Miss. River?	56,632
1943 (?)	East of Miss. River?	7,270
Summer 1944	Sister Lake	29,185
Summer 1944	Lake Felicity	29,022
Summer 1945	Bay DuChene	15,251
Summer 1945	Lake Tambour	16,383
Summer 1945	Sister Lake	19,825
Spring 1945	Bay Boudreaux	40,965
Summer 1946	Bay DuChene	20,048
Summer 1946	Sister Lake	19,213
Summer 1946	Bay Boudreaux	40,000
1948	Mississippi Sound; West of 3-mile Bayou	19,014
1948	Bell Pass (seed)	10,618
1949	Sister Lake	21,560
1949	Lake Felicity	17,539
1949	Mississippi Sound; Turkey Bayou	14,314
1949	Bayou LaMer (seed)	8,255
1950	Sister Lake	34,620
1950	Lake Felicity	17,042
1950	Bay Gardene (seed)	14,475
1950	Bay Gardene (seed)	12,335
1951	Sister Lake	40,057
1951	Lake Felicity	10,000
1951	Bay Gardene (seed)	10,650
1952	Sister Lake	51,117
1952	Half Moon Island	35,841
1953	Sister Lake	30,474
1953	Half Moon Island	29,817
1954	Half Moon Island	36,205
1955	Mississippi Sound; Bay Boudreaux	34,883
	Total	1,653,823

the summer of 1944, and that later, in 1949, 1950, and 1951, additional large plantings were made. At considerable expense, a watchman's camp was erected on the west shore of the lake. But fungus disease and conchs proved too much, and the project was allowed to die. Walters (1952) stated, "Lake Felicity shell plantings to date have been unsuccessful."

The use of Sister Lake (Caillou Lake in Fig. 21) as a seed reservation began very early. Its history is probably better known than that of most areas, and it appears to have had a long, relatively useful life in seed oyster production (Table 6). Payne made the first plantings of shell in 1917. Sister Lake produced good oysters with intermittent

TABLE 6

Sister Lake shell plantings, 1917 to 1953

Year	Amount planted barrels
May 1917	16,238
Summer 1941	50,000
Summer 1943	91,632
Summer 1944	29,185
Summer 1945	19,825
Summer 1946	19,213
1949	21,560
1950	34,620
1951	40,057
1952	51,117
1953	30,474

periods of poor condition up to 1941, when a regular system of shell plantings was developed. From 1943 each year saw from 19,000 to 90,000 barrels of shell bedded in Sister Lake. The amounts averaged 37,520 barrels per year from 1941 to 1953 inclusive. Shells are bedded at a rate of about 250 barrels per acre. This would mean that about 150 acres were shelled each year. Sister Lake contains about 7600 acres; thus each year about two percent of the area of the lake was covered. A gain in acreage covered would be made for about three years, so that an estimated six percent of the entire area of the lake was maintained in a shelled condition at any one time. Payne (1920), in a map of the Louisiana coast, showed the entire bay as a prolifically producing area. It is not believed that more than one-fourth has ever been productive. St. Amant *et al.* (1956) estimated that only about 600 acres is now productive, all in the north end of the lake. Such bays usually have large areas of mucky bottom in the center and only the margins and some central ridges are hard enough for good production. Perhaps about 75 percent of the productive area has been kept shelled for the past 15 years; if so, this is a very good record. Recently high-salinity water has encroached on Sister Lake. St. Amant (1955) showed that between 1951 and 1955 those months in which salinity was high enough to support drills increased in number from two per year to ten. He recorded increasing fungus disease as well as increase in conchs, and stated that the Bay was 90 percent useless in 1955. Reclamation of this once great oyster-producing bay can be accomplished only by reduction of salinity either through cyclic weather change or through introduction of fresh water by engineering methods. It would require an enormous volume of fresh water, estimated at about 1.6 billion cubic feet daily, to dilute nine square miles of high-salinity sea water.

With the exception of several such areas as Sister Lake, it is believed that state shell planting in Louisiana is more of a public relations practice than anything else. The acreage shelled is not impressive when compared with the total area of natural reefs. McConnell (1932) stated that there were 50,000 acres of natural reefs in St. Bernard and Placquemines Parishes east of the Mississippi. From 1929 to 1953, a quarter century, an average of 57,248 barrels per year of shells was planted. At 250 barrels per acre this would plant 229 acres annually or less than 0.25 percent of the natural reef area east of the Mississippi alone, if it were all applied there.

Physical and Chemical Conditions

Louisiana's oyster-producing areas are shown on the map, Fig. 5. Currently the best

producing area is the "Louisiana Marsh" in St. Bernard Parish east of the Mississippi River. Some of the St. Bernard Parish production is in Mississippi Sound. The great productivity of this section depends on the Pearl River which keeps salinity low in the adjacent part of Mississippi Sound and the more northerly part of the St. Bernard marshes. In Placquemines Parish, east of the Mississippi, oysters are produced as far south as Grand Bay; most of this area from American Bay east to Black Bay and south to Quarantine Bay contains large seed reefs. Salinity in eastern Placquemines bays is controlled by the so-called Bohemia (or Point a la Hache) Spillway, actually a gap in the levees on the east bank of the Mississippi beginning just south of Point a la Hache and extending about five miles down-river. Silting has much reduced the effectiveness of this spillway in recent years.

Some oysters of low grade grow all the way around the end of the Mississippi River. These are in the marginal bays which are sufficiently open to the Gulf to let oysters maintain themselves intermittently. In flood years all of the oysters may be destroyed. These bays are now insignificant in overall production, but in the early years of the industry they were of considerable value.

On the west flank of the Mississippi River, the productive area begins at Bay Tambour and Sandy Point Bay near the edge of the freshwater marsh. From there oysters grow in all the bays and bayous to a point above Bay San Bois, westward to Little Lake (northwest of Barataria Bay proper) and to Bayou Lafourche. Most of this area has high salinity and high oyster mortality, as a rule. It is the locale of the major part of the leases on which suits were based. West of Bayou Lafourche, important oyster production extends to the Atchafalaya River, and includes Timbalier and Terrebonne Bays and all tributary bays and bayous. Around Sister Lake (Caillou Lake) and Bay Junop is another intricate network of small bays and bayous, probably the most satisfactory of the producing areas of the state. Even this seems to be deteriorating because of the intrusion of high-salinity water. West of the Atchafalaya River there is some production, and oysters grow intermittently all the way to the Sabine River. In Atchafalaya Bay proper are the most extensive natural reefs in the state (Payne, 1912). These are mostly useless because they are solid high reefs, thickly studded with small misshapen oysters which are so tightly packed and attached that they can be gathered only with a crowbar. Most of the Point au Fer reef near the mouth of the Atchafalaya is dead (from low salinity) and is being mined for shell. The westward end of the Point au Fer reef extends to Marsh Island where extensive reefs on the Gulf side are dredged for seed.

An understanding of this complex of marshes, bayous, and bays depends on an understanding of the nature of deltas of large rivers. The Mississippi River follows a channel between ridges of its own building, the natural levees, on which are built the artificial ones. The large bayous are deltaic distributaries of the Mississippi, or once were. These also follow ridges, their natural levees. Except for the Atchafalaya River and the "passes" in the lower subdelta, these distributaries have been severed from their connections with the Mississippi by man-made levees, and no longer serve as distributaries. Between the major bayous are topographic basins, the deepest parts of which are the major bays. Flanking the major bays on either side are smaller satellite bays, and others of these extend inland. These inland bays may be far enough removed from the Gulf to contain fresh water only. An intricate system of tidal drains joins major and minor bays to each other. Many of these are sections of relict bayous which originally were parts of the Mississippi distributary system.

The Barataria basin, lying between Bayou Lafourche and the Mississippi, is by far the most important basin in regard to the number and magnitude of claims of oyster mortality in 1940–1947. For this reason it was studied most closely by Research Foundation personnel and will be described in detail in this section.

A less well-defined basin is that containing Timbalier and Terrebonne bays and their satellite bays and bayous. Originally (as late as 1860) this basin was at least three separate basins. The first extended from Bayou Lafourche westward to Terrebonne Bayou, which at that time occupied a raised ridge which came to an end only a few miles from the Gulf near Caillou Island. The second included Terrebonne Bay and lay between Bayou Terrebonne and Bayou Petit Caillou. The third was west of Bayou Petit Caillou. At the present time, the lower ends of Bayou Petit Caillou and Bayou Terrebonne are drowned in the expanding and deepening bays and the area is one large basin, indefinitely segmented in the northern portion.

Bayou Lafourche has its origin at Donaldsonville, some 75 miles as the crow flies above its mouth just east of Timbalier Bay and west of the barrier islands which separate Barataria Bay from the Gulf. It is approximately 100 miles long. The Mississippi, forming the eastern boundary of the Barataria basin, is much longer, taking a circuitous route from Donaldsonville to the sea. Along the axis of the long triangular basin contained between these two major waterways are several large lakes, beginning at the northwest end with Lake Des Allemands, which is fresh water, and ending on the southeast with Barataria Bay. The map, Fig. 6, shows numerous distributaries radiating from Bayou Lafourche and the Mississippi which lead into small tributary bays which in turn connect with Barataria and Caminada Bays. Most of these minor distributaries, severed from their parent distributaries by levees, and blocked in the fresh water zones with water hyacinth and alligator weed, no longer carry fresh water into the bays in significant amounts.

The eastern margin of the Barataria basin continues down to the west side of the active delta of the Mississippi River. The river is no longer depositing sediment outside of its bed except in the area of fresh-water marsh forming the last 25 miles of the end of the delta.

Across the southeast end of the Barataria basin there is a chain of barrier islands. The principal islands are Grand Isle and Grand Terre, which, with those flanking to the east and west, form a large arc separating the basin from the Gulf. The sea rim extending from Bayou Lafourche to the active Delta is broken by several passes, of which Barataria Pass is the largest.

All of the oyster-producing areas of the Barataria basin are in the marshlands (including the bays). Variations in elevation are so slight that they are measured in inches rather than feet. Even the natural levees along the chenieres and major bayous are only inches higher than the marsh, and depths measurable in feet occur only in the bays and bayous.

The coast of Louisiana, especially the active Delta, is sinking at a fast rate. Gunter (1952) has discussed references and rates of settling. According to his summary, the rate varies from 0.05 to 0.163 foot per year. This includes both actual land subsidence and relative sinking due to rise in sea level. Marmer (1954) records a rise in Gulf level at Pensacola of about 0.3 foot in the decade from 1940 to 1950, and at Galveston the rise was 0.5 foot. This indicates a rise of about 0.4 foot at Grand Isle, accounting for about one-third of the average change.

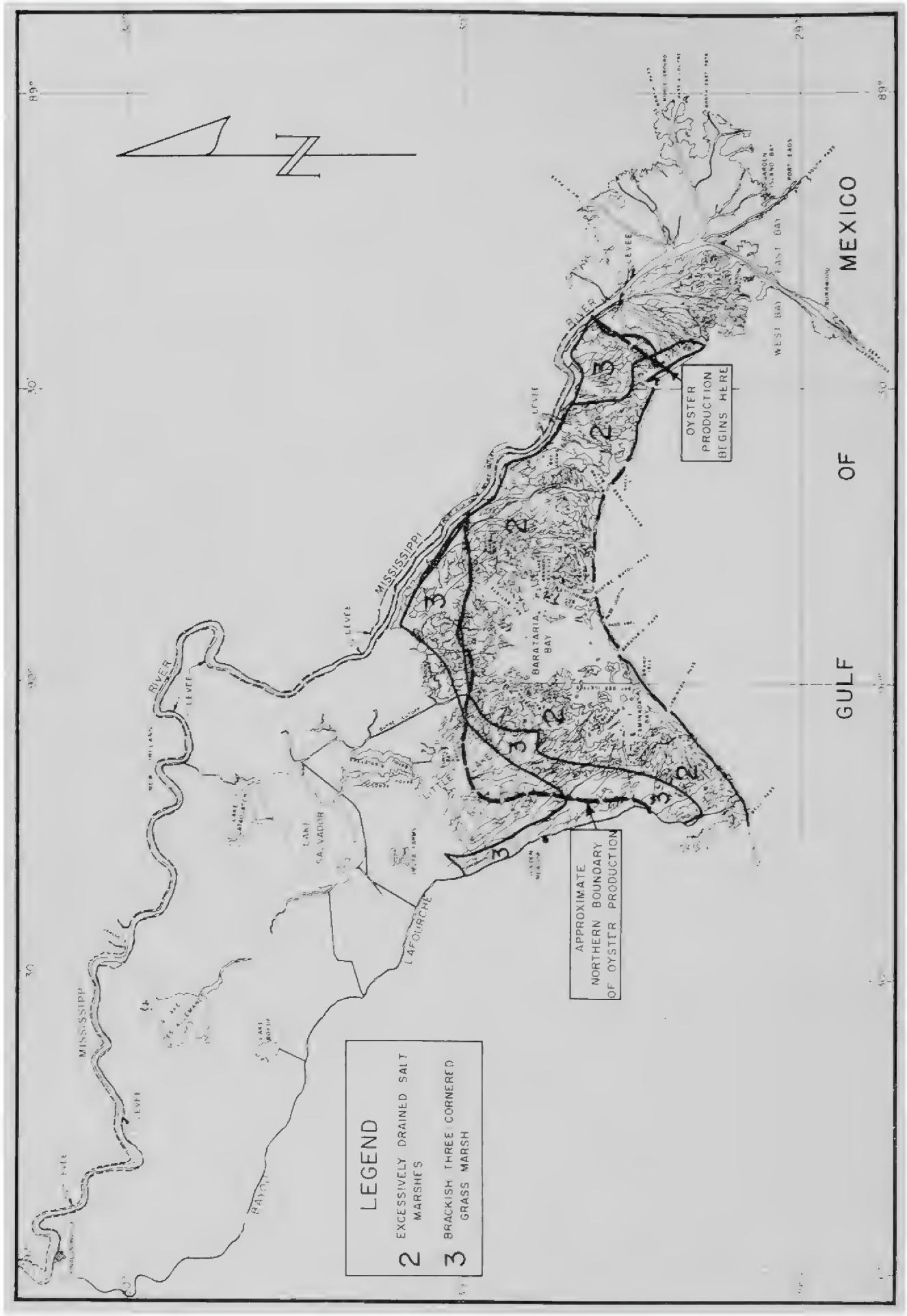

FIG. 6. Map showing the Barataria Bay basin, with its drainage area between Bayou LaFourche and the Mississippi River. The highest mortalities of oysters occur in the southern two-thirds of the salt marshes shown on this map.

Coupled with the sinking are other changes of a far-reaching nature. Disintegration of the shore-lines of bays and of the barrier islands is proceeding at a fast rate. Scour due to increased fetch of the wind (Price, 1947) has prevented silt accumulations, so that the mean depth of bays is increasing. Levees of the Mississippi prevent large de-

posits of silt in the delta flanks which under natural conditions would be expected in flood times. The exotic plants, water hyacinth and alligator weed, are producing important changes in the ecological complex. The increase in the efficiency of the Mississippi River levees is one of the most important of the changes, because it has eliminated crevasses in flood period. The date of the last important crevasses which let river water into the Barataria region (1927) is an important turning point for the oyster industry.

Rasmussen and Lynch (1949) have shown that sediments in Barataria Bay are becoming finer, and that there was a tendency toward poorer sorting of particle size in 1949 compared with data from 1934 (Krumbein and Aberdeen, 1937). It is believed that more frequent encroachment of high-salinity water in recent years tends to precipitate finer particle sizes by flocculation. Other changes possibly contributing to deposit of finer sediments are a decrease in frequency of scouring by floods from crevasses, a decrease in tidal current rates in certain parts of the bay due to closure of passes to the east of Grand Terre, and acceleration of the rate of disintegration of shorelines. The last change would produce a greater deposit of unsorted materials due to slumping. Barataria Bay depths seem not to be in equilibrium with the width and length of the Bay. There may be a lag in restoring equilibrium following abrupt changes caused by the improvement of levees and the severing of major distributaries.

Nearly all of the oyster production of the Barataria basin is in the "excessively drained salt marshes" of O'Neil (1949). However, as salinity has increased, the oyster industry has pushed into Little Lake northwest of Barataria Bay proper, and elsewhere into the "floating marsh" areas (Fig. 6). O'Neil's areas, which he believes are ecological entities, are shown on this map. Certainly the greatest part of the oyster industry of the Barataria basin is contained in the excessively drained marsh zone, all of which is subject to tidal currents. Our own ecological studies were for the most part directed toward analysis of the salinity patterns, water movements, and other major environmental factors affecting oysters in that zone.

Tides and Tidal Currents

Reports by Marmer (1935, 1947, 1948) contain data on volumes of water movement through the principal passes into the Greater Barataria Bay area and Timbalier-Terrebonne Bays, as well as data on the tides and tidal currents. A tide station has been maintained since 1947 in Bayou Rigaud, an arm of Barataria Bay near Barataria Pass, in collaboration with the U. S. Coast and Geodetic Survey. During 1947 two subsidiary stations for collecting tide data were maintained at Manilla Village in Upper Barataria Bay and in the northern part of Bay Baptiste, a northeast extension of Barataria Bay.

Marmer (1954) described the general features of tides in the northern Gulf area, and defined the types of tides, seasonal variations in tide level, and cyclic increase in Gulf level from 1909 to 1950.

Tides are of the diurnal type, i.e., there is one high and one low per day. There are occasional days when two high water periods and two low water periods occur in one day. Times of high and low water fluctuate erratically from day to day, but tend to average out, over a long period, about 50 minutes later each succeeding day. At periods of equatorial tides the range may be only 0.1 foot or even zero on rare occasions. Such low ranges result from the effect of wind. The range at equatorial tide periods is usually 0.2 to 0.4 foot. At tropic tide periods normal ranges of 2.2 feet have been recorded, but

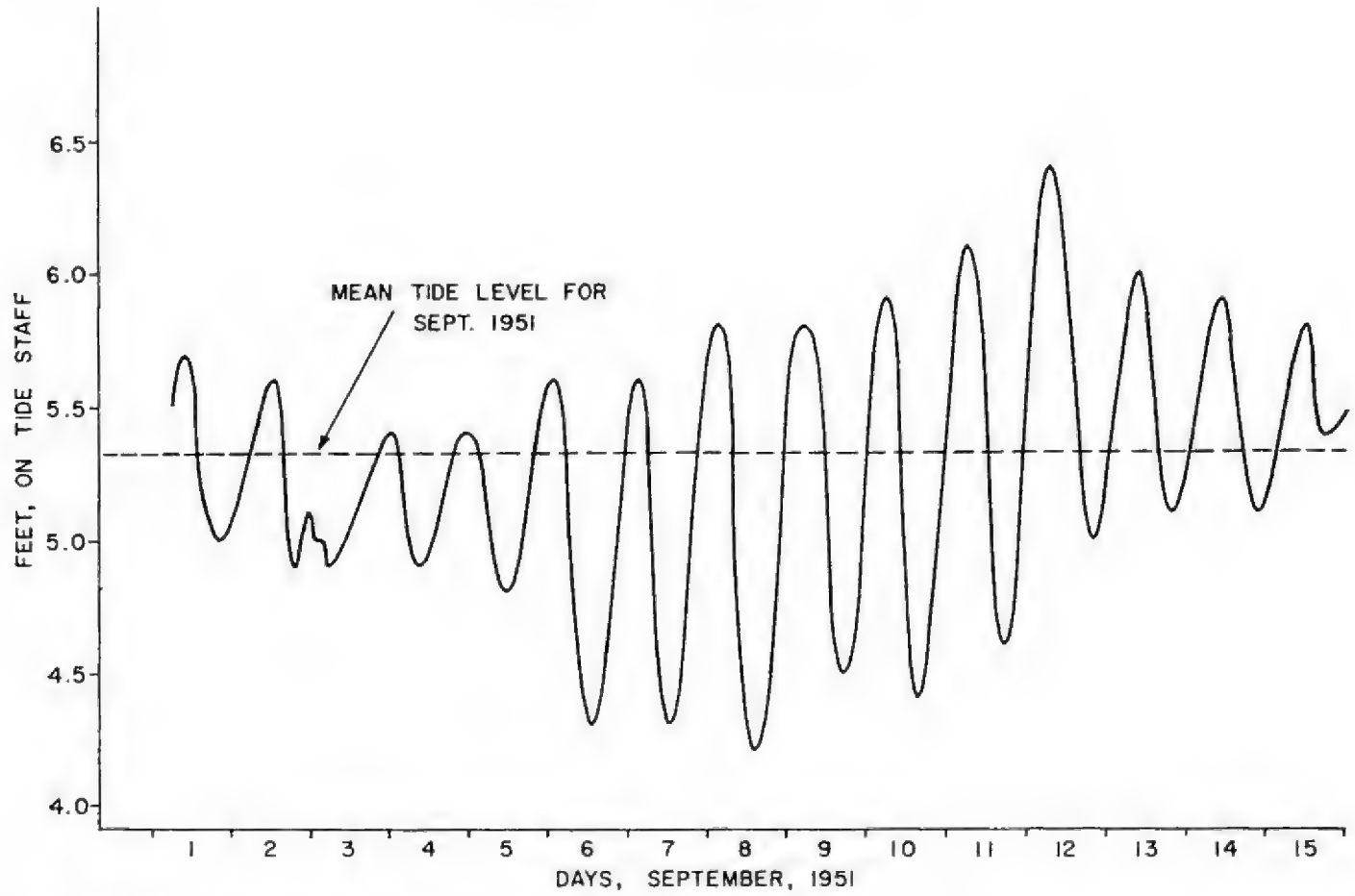

FIG. 7. Typical tides at the Bayou Rigaud station from an equatorial tide period through a tropic tide period, and return, Sept. 1 to 15, 1951.

the usual range is 1.4 to 1.8 feet. Fig. 7 shows tides for the period from September 1, 1951 to September 13, 1951; this graph may be taken as typical of a period of high mean tide level.

In Fig. 8 is shown a graph of tides before, during, and after the passage of hurricane Flossie in September, 1956. The peak shown on September 24 is the highest level attained in the period from 1947 to 1957. Damage to roads and property by the

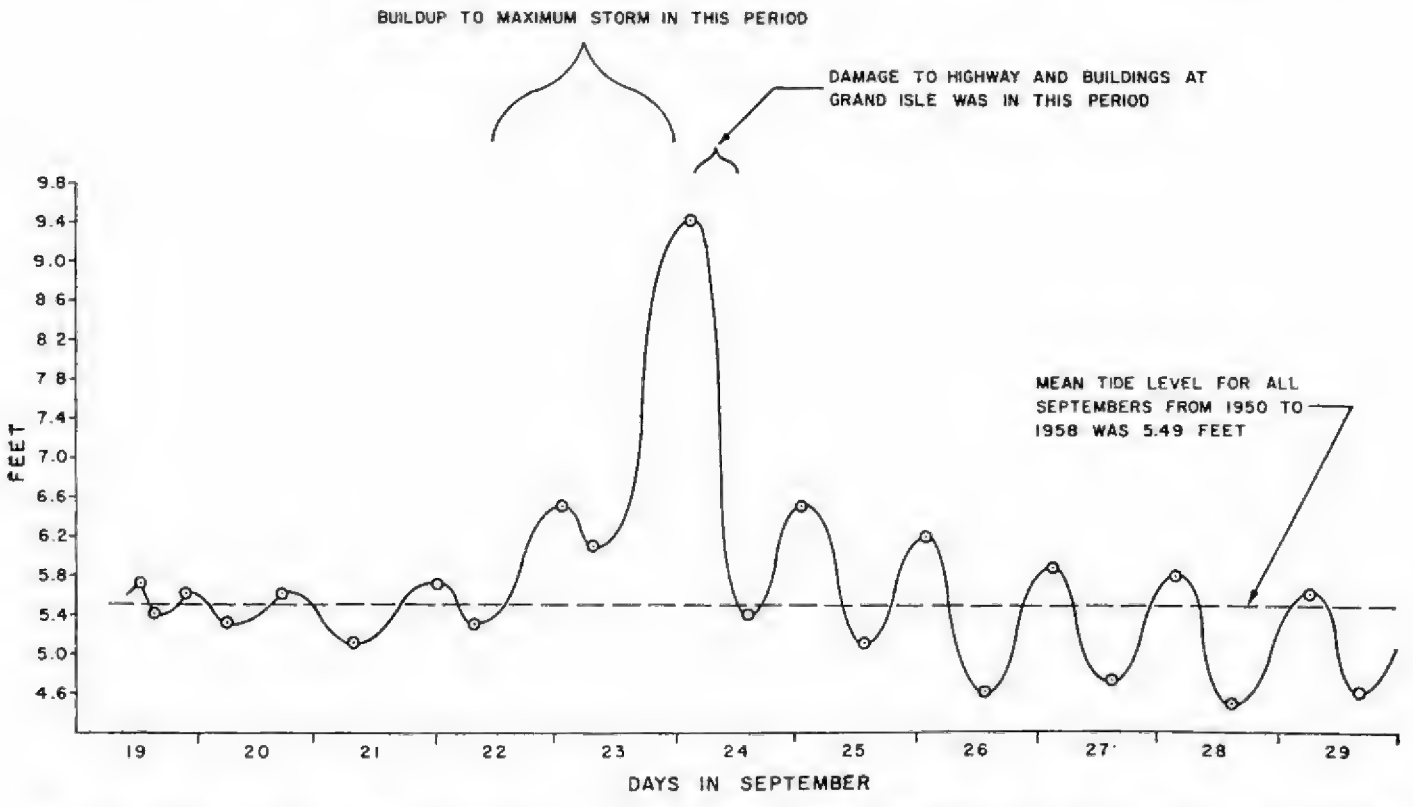

FIG. 8. Tides at Bayou Rigaud during passage of hurricane Flossie, data from Sept. 19 to Sept. 29, 1956.

receding tide following the passage of the hurricane was very great; little damage occurred as the tide rose. Winds in this hurricane were not exceptionally high. The period of increasing wind velocity coincided with a normal flood tide, and it happened to be near the maximum of a tropic tide period in the month with the highest mean tide level of the year. This coincidence accounts for the great build-up of Gulf water in the bays and marshes behind the barrier islands, Grand Isle and Grand Terre. When the wind changed with passage of the hurricane, it coincided with the beginning of ebb tide. The consequent rush of waters back into the Gulf produced great damage to highways, boats, and buildings.

Mean tide levels fluctuate in a regular rhythm throughout the year. The lower mean levels are in January, and the highest in September (Marmer, 1954). Fig. 9 shows the mean monthly tide levels for the six-year period from 1951 to 1956 as recorded by the Bayou Rigaud tide gauge.

The increase in mean tide level from August to September is of the greatest significance in its effect on salinity levels in the bays and particularly in the marshes far above the bays. During approximately one month's time the bays may deepen by more than one-half foot. With low rainfall in the marsh drainage area, high evaporation rate due to midsummer temperatures, and high transpiration rate of lush summer marsh growth, the net flow of water from Louisiana bays during this month is not to the Gulf but into the marshes. Much of the marsh area is near sea level and large areas are covered with floating marsh. Transpiration can, under conditions of rainfall deficit, actually reduce marsh level at the same time that tide level takes a steep climb. The effect is a rapid transfer of salt water far inland. A more detailed discussion of this effect is contained in the section on salinity.

Range of tide also varies over a cycle of 18.6 years (Marmer, 1954). The maximum-range periods were in 1931–32 and in about 1950–51, while the minimum-range period between these two peaks lies at about 1941, with another to appear in 1960.

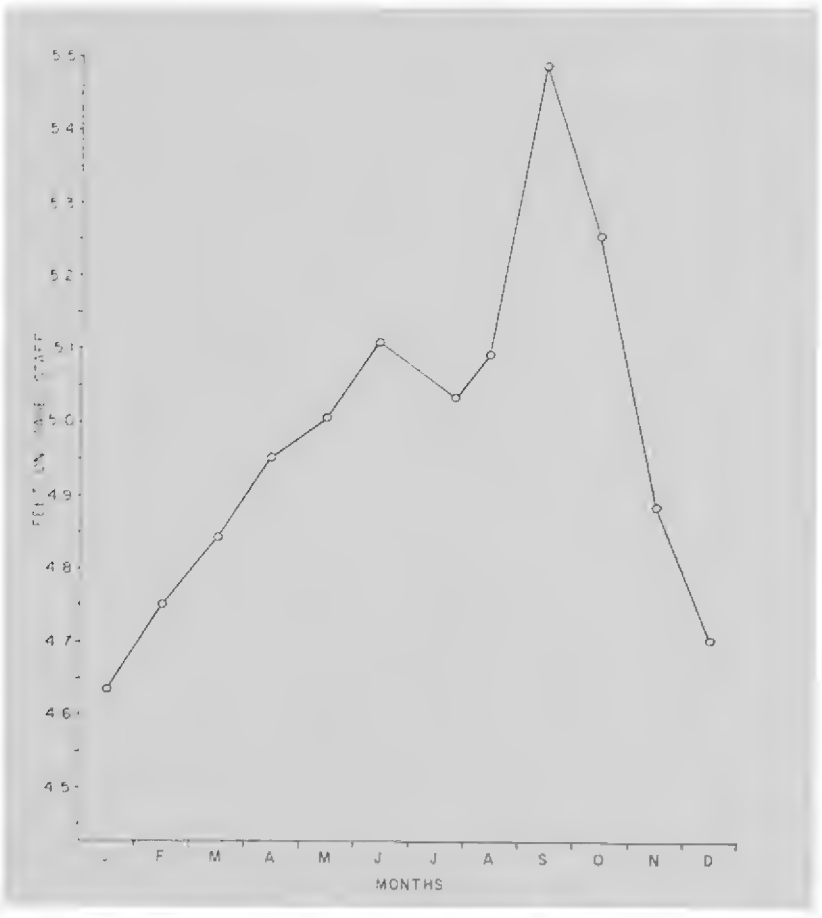

FIG. 9. Mean monthly tide level at Bayou Rigaud, 1950 to 1958.

Perhaps the most important factor related to tides has been a steady rise in sea level over most of the century, affecting the entire Gulf. According to Marmer the amount of this rise at Pensacola was about 0.01 foot per year from 1924 to 1941. Thereafter the rate was sharply increased to 0.03 foot per year. At Galveston the increase in this latter period was at a rate of 0.05 foot per year. Study of tide records at the Bayou Rigaud station for the years 1951 to 1956 indicates that the rise is continuing, the rate approximating that at Pensacola. Whether the rise in sea level is an actual rise, a relative rise due to sinking coast line, or both, is not clear. It is believed to be a combined effect of both. The result has been a general encroachment of saline waters inland, the effects of which are discussed elsewhere. Marmer's figures showing rise of Gulf level at Pensacola and at Galveston are reproduced in Fig. 10.

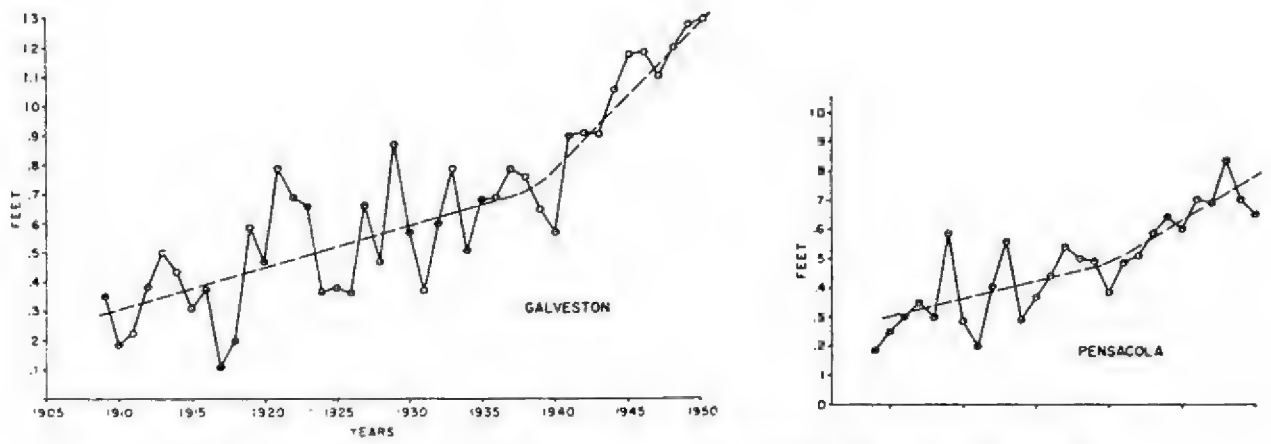

FIG. 10. Yearly sea level at two Gulf stations, after Marmer, 1954.

Marmer (1935) estimated the area of tidal waters of Timbalier and Terrebonne bays as 450 square miles in round numbers. From tidal ranges in the lower, middle, and upper parts of the bay, and the Greenwich time intervals, he calculated that an average of about 15 billion cubic feet of Gulf water flowed through the passes into these bays on each daily flood tide. At tropic tide periods the amount was more, and at equatorial tide periods it was less.

Similarly, he calculated the movement into Greater Barataria Bay through the several passes (Caminada, Barataria, Quatre Bayou, and Abel). The data are reproduced here as Table 7 which is taken from Marmer (1948).

TABLE 7

Tidal flow of water through passes of the Greater Barataria Bay Area. Figures are millions of cubic feet. Data from Marmer (1948)

Pass	Flood flow	Ebb flow	Ebb excess
Caminada	627	653	29
Barataria	3229	3438	209
Abel	129	212	83
Quatre Bayou	874	1005	131

Data on currents from which Marmer calculated the volumes in Table 7 were obtained during August, 1947. Those for Barataria Pass are most reliable, observations having been carried through an entire tidal cycle. Austin (1955) used Marmer's data to calculate a flushing time of 23 days for all of Barataria Bay. The total mean volume of water in Barataria Bay was calculated by Marmer to be about $15{,}391 \times 10^6$ cubic feet.

Currents in Barataria Pass had a mean flood strength of 0.79 to 1.55 knots and mean ebb strengths varied from 0.87 to 1.59 knots. In upper Barataria Bay, in open water, mean velocity was 0.21 knots at flood and 0.31 knots at ebb tide. These latter figures are representative of open bay current conditions where cross-sectional areas are very large.

Oxygen, Redox Potential, and Hydrogen Sulfide in the Barataria Area

Oxygen in Barataria Bay was studied by Hewatt (1949, 1953, 1955) and Jensen (1949). Most of the oxygen determinations were made in 1948 by Jensen and associates. Oxygen measurements were made on more than 1100 samples taken from all parts of the Greater Barataria area in all months of the year. Water samples were taken from a level about one foot over the bottom, and simultaneous measurements of redox potential (Eh) were made. The relation of the readings to tidal cycle was determined. The relations of oxygen tension and redox potential were also studied by taking continuous Eh readings accompanied by oxygen titrations over short periods at set stations. Hewatt made supplementary studies on conditions in small marsh ponds distributed from brackish to high-salinity areas.

Many water samples were tested for hydrogen sulphide. The number of positive readings was very low; in fact, in the open parts of Barataria Bay they were almost non-existent. Positive tests could be found occasionally in some small bayous and shallow marsh ponds where concentrations of organic substances were high. Bleedwaters were also checked for hydrogen sulphide and an attempt was made to measure the influence of these in contributing hydrogen sulphide to the waters. This influence was found to be too small to measure.

Oxygen determinations by Jensen were made by a modified Winkler method. A Beckman Model G meter was used for measurements of oxidation-reduction potential; the results were expressed in millivolts. Platinum and calomel electrodes were used, and readings were made in the field. Except where noted all determinations were at a level of about one foot over the bottom.

Fig. 11 is a summary of 1134 values for oxygen taken in all months of 1948 and

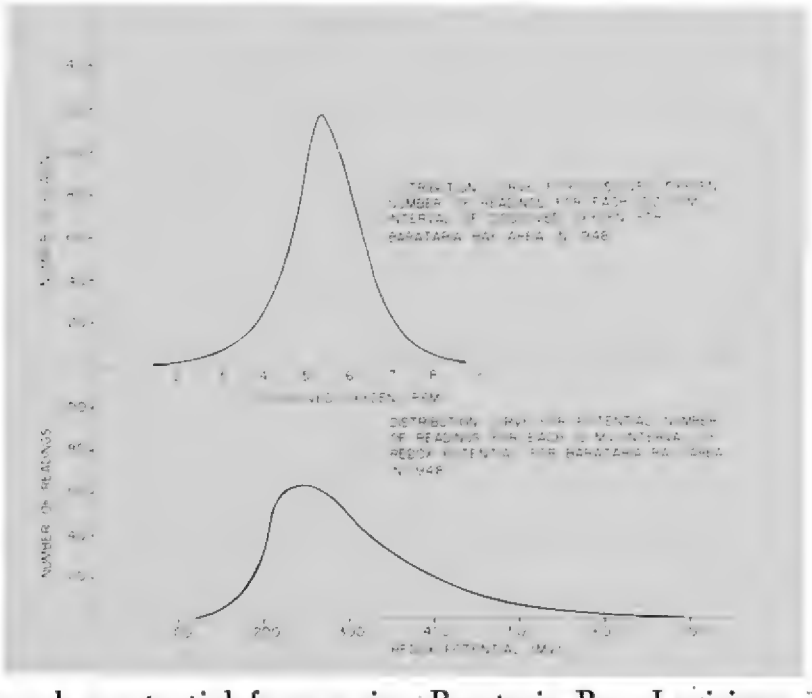

FIG. 11. Oxygen and redox potential frequencies, Barataria Bay, Louisiana. 1134 determinations 1948.

generally distributed over Barataria Bay. The same figure presents a summary of 1134 simultaneous determinations of Eh (Jensen, 1949). About 43 percent of all oxygen determinations fell between 5 and 6 ppm. Only three titrations showed oxygen tension below 2 ppm and none below 1.5 ppm. No determinations showed supersaturation. Only 24 (2%) of 1134 determinations showed less than 50% saturation. The oxygen content of samples taken on ebb tide averaged slightly higher (5.44 ppm) than that of samples taken on flood tide (5.16 ppm). This is probably a reflection of a difference in salinity.

A study of the distribution of values of oxygen by months shows that there was a tendency toward slightly higher readings in the winter months, but the difference is not as much as expected. In the summer when mortality rate of oysters was high, oxygen values clustered around the 5 ppm mark. Most of the water samples in which the oxygen content was below 50 percent of saturation were taken in the period from April to August.

It was established that oyster mortality rates were significantly higher in lower Barataria Bay than in the upper reaches of the bay. On the average, mortality rates at the Sugar House Bend station, just to the north of Grande Terre Island, were about 2½ times the average rates in Bay Chene Fleur, above Barataria Bay proper. It seemed reasonable to assume that if low oxygen was a factor in these mortalities, the oxygen determinations would be significantly lower in the lower bay areas. The redox and oxygen values were accordingly rearranged to show comparative distribution of values for upper, middle, and lower Bay areas. The data are presented in Fig. 12. Inspection of the graphs shows that there is no significant difference in the distribution of oxygen values in the three areas. Oxidation tendencies are shown to be slightly higher in middle and lower bay, with a tendency to formation of a bimodal curve for redox potential in the lower bay; the lower of the two modes shown for the lower bay is absent from the upper bay graph, but shows weakly in the middle bay values.

As expected, there is a broad general relation between the oxygen values and the oxidation—reduction potentials. This relation was checked further by means of a series of simultaneous measurements in three areas over a number of hours. Six such comparisons were made and the data are presented in Fig. 13. Four of the "runs" were made in low-salinity water in the Texas Company's LaFitte Oil Field during December, 1947, and January, 1948. All of these were made in nearly stationary waters of the small boat canal system, into which flow large amounts of oil bleedwater. The other readings were taken in middle Barataria Bay at the Bassa Bassa Station, and at Station 51 in mid-lower Barataria. The latter station was at a tank battery of the Texas Company which at the time was producing an insignificant amount of bleedwater.

Three of the runs at Camp LaFitte show extraordinary amounts of oxygen, up to 8.8, 9.0, and 10.6 ppm, probably from plankton blooms. Under laboratory conditions it has been observed that oil bleedwater produces such blooms. Owen (1955, section 6) presented data showing that oil in water greatly enhances growth of diatoms and other chlorophyll-bearing organisms. For a contrary view see Galtsoff *et al.* (1935). The fourth run showed a low initial concentration of oxygen which increased to more than 6 ppm in 14 hours, the greater part of the increase being in daylight hours. All Eh readings were positive values and showed that the high oxygen values influenced similarly high positive redox potential. Minor independent fluctuations of oxygen values and redox values occurred when both were high.

At Station 51 redox potential values apparently fluctuated through a wide range with

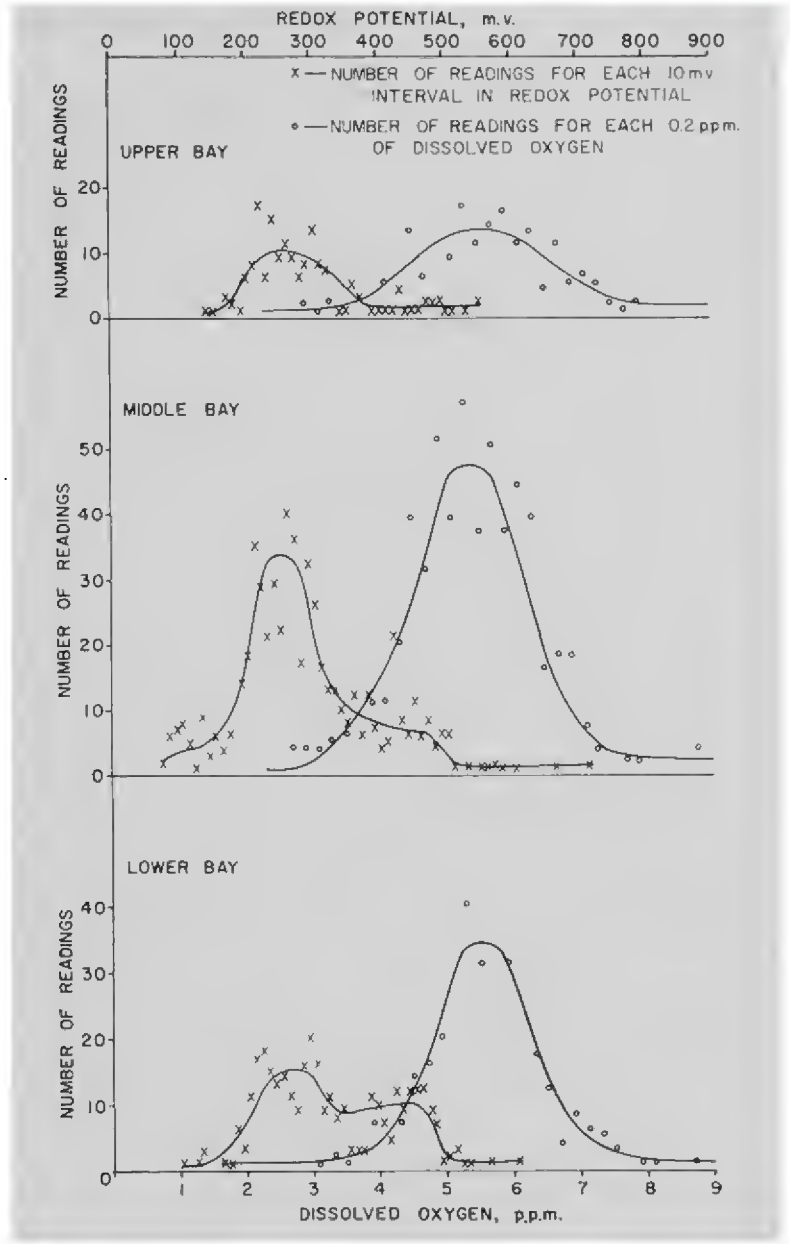
DISTRIBUTION CURVES FOR DISSOLVED OXYGEN & REDOX POTENTIAL IN BARATARIA BAY AREA IN 1948
REDOX POTENTIAL, m.v.
0 100 200 300 400 500 600 700 800 900
x — NUMBER OF READINGS FOR EACH 10 mv INTERVAL IN REDOX POTENTIAL
• — NUMBER OF READINGS FOR EACH 0.2 ppm. OF DISSOLVED OXYGEN
UPPER BAY
MIDDLE BAY
LOWER BAY
NUMBER OF READINGS
DISSOLVED OXYGEN, p.p.m.

FIG. 12. Comparison of values of oxygen and redox potential for upper, middle, and lower Barataria Bay.

no corresponding oxygen changes. Here there were evidently reducing agencies which influenced the redox independently. At Bassa Bassa Bay, fluctuations of Eh were minor as were those of the oxygen values.

Redox potential values in Figs. 11, 12 and 13 show that a wide range of positive values occurred in the waters, with no negative readings. Some were phenomenally high and there were rare values in excess of +700 millivolts. On the other hand, Jensen and co-workers found that Eh readings became negative whenever the electrodes entered bottom muds.

Valentine and McCleskey (1956) have presented some interesting data on the redox potential values in Barataria Bay. They show a generally higher redox potential near the bottom than occurs at the surface of the water, which they suggested might be due to concentration of algae near the bottom. All of their values were positive but the range was lower than shown by Jensen's measurements. Valentine and McCleskey made a relatively small number of measurements, confined to the summer months. They thought that organic matter in the bottom influenced the redox potential, a conclusion which seems to be correct. Our own studies have shown that the smaller bays are relatively rich in plant debris as compared with the large bays, and that the carbon content of

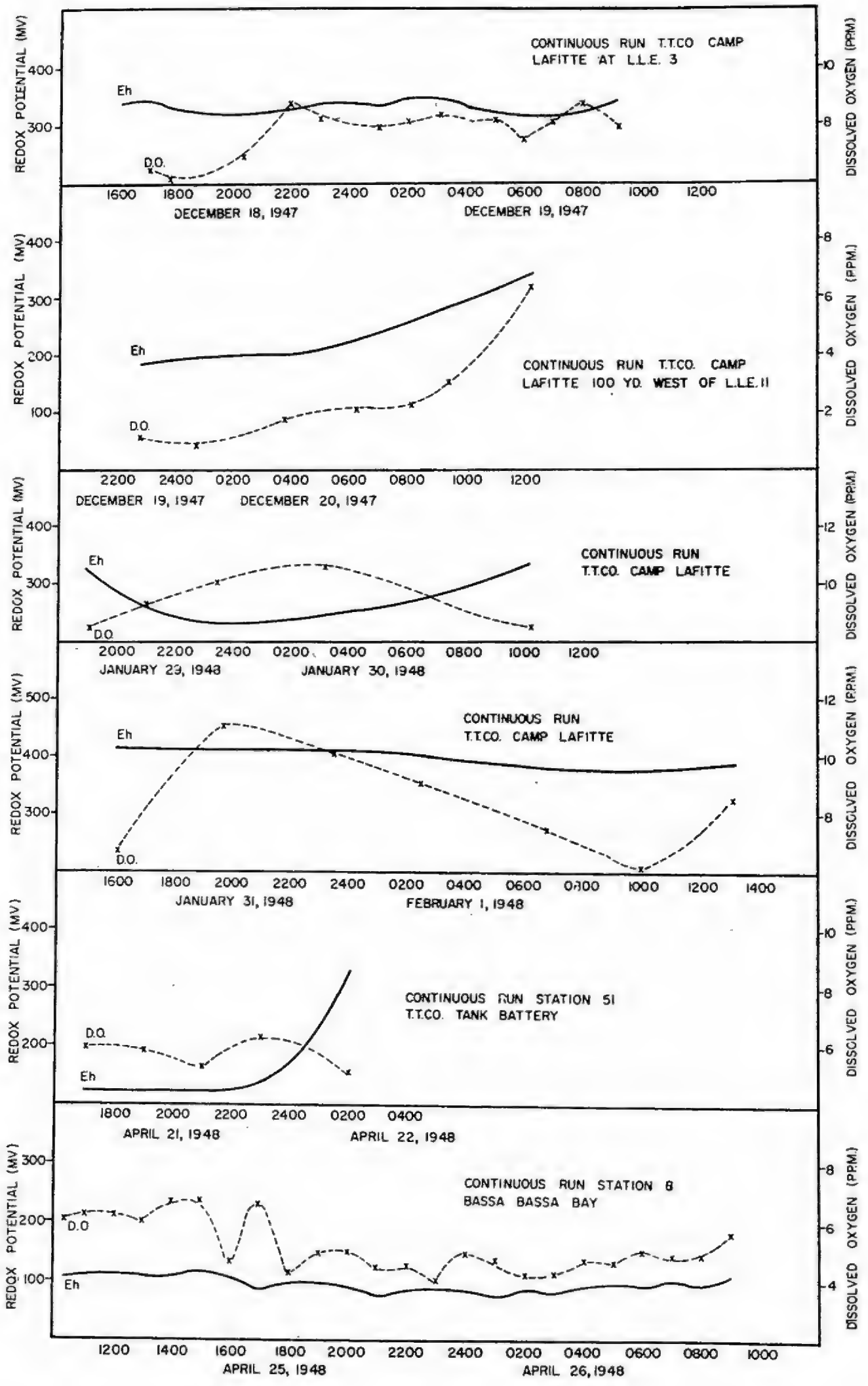
CONTINUOUS RUN T.T.CO CAMP
LAFITTE AT L.L.E. 3
Eh
D.O.
REDOX POTENTIAL (MV)
DISSOLVED OXYGEN (PPM.)
1600 1800 2000 2200 2400 0200 0400 0600 0800 1000 1200
DECEMBER 18, 1947
DECEMBER 19, 1947
CONTINUOUS RUN T.T.CO. CAMP
LAFITTE 100 YD. WEST OF L.L.E. 11
2200 2400 0200 0400 0600 0800 1000 1200
DECEMBER 19, 1947
DECEMBER 20, 1947
CONTINUOUS RUN
T.T.CO. CAMP LAFITTE
2000 2200 2400 0200 0400 0600 0800 1000 1200
JANUARY 29, 1948
JANUARY 30, 1948
CONTINUOUS RUN
T.T.CO. CAMP LAFITTE
1600 1800 2000 2200 2400 0200 0400 0600 0800 1000 1200 1400
JANUARY 31, 1948
FEBRUARY 1, 1948
CONTINUOUS RUN STATION 51
T.T.CO. TANK BATTERY
1800 2000 2200 2400 0200 0400
APRIL 21, 1948
APRIL 22, 1948
CONTINUOUS RUN STATION 8
BASSA BASSA BAY
1200 1400 1600 1800 2000 2200 2400 0200 0400 0600 0800 1000
APRIL 25, 1948
APRIL 26, 1948

FIG. 13. Comparison of redox potential and oxygen values at six stations in the Barataria Bay area. Samples were taken simultaneously.

muds near shore is greater than in muds at considerable distance from shore. Slumping, as the shorelines disintegrate, causes decaying marsh turf rich in plant debris to accumulate along shores. Examination of the locations of Valentine and McCleskey's redox potential values in the low positive class shows that they are all in the smaller detritus-laden bays or near disintegrating shorelines.

Hewatt (1953b) has shown that the oxygen content of water in small marsh ponds may change very rapidly. One pond studied by him recovered from a low of five percent saturation to a high of 112 percent in the period of 10 hours. The minimum value was recorded at 0415, the maximum at 1430, showing the effect of photosynthesis. Part of the gain appeared to be the result of an inflowing tide. This pond was only six inches to one foot deep, depending partly on the tide stage. Such ponds lack the conservatism of the large bays, in capacity to limit oxygen fluctuation. The pH changes in this pond were rather small during the extreme oxygen fluctuations recorded above, indicating that redox potential may not have changed greatly, pH being to some extent a function of the Eh (ZoBell, 1946).

Turbidity

Bays and bayous of the Mississippi River delta depressions are generally turbid. Much of this turbidity is due to materials carried to the Gulf by the river in the past; because of the levees the present suspended silt load of the river is a relatively minor factor; the saline Gulf water, into which the river now passes, precipitates most of its silt load. However, there are no oyster-producing areas in Louisiana where the water is clear in the sense that offshore waters are clear. The normal turbidities occurring in Barataria and adjacent bays are always high, probably higher than in most other oyster-producing regions of the United States. When the investigation began, there seemed to be a chance that excessive turbidity might in some manner affect mortality rates of oysters in Louisiana.

Data on normal turbidities in Barataria Bay, collected by W. G. Hewatt, have been presented in serveral reports (Hewatt, 1949, 1953a, 1955). Most of Hewatt's turbidity measurements were made by means of photoelectric cells set at a point on a glass tube through which sea water was pumped in a constant stream. Hewatt estimates that about 100,000 hours of turbidity readings were recorded on Esterline Angus recorders. Calibration was made by means of standard fuller's earth suspensions, and checked against a Jackson candle turbidimeter. The stations for taking turbidity measurements were the same as those for salinities (see Hewatt, 1951, for a complete series).

A combination of high wind and rainfall operated to produce the highest turbidities recorded. Some of these were in the range of 400 to 900 ppm. Such high turbidities were of short duration, normally lasting a matter of hours. But turbidities of several days duration were encountered in the range of 200 to 400 ppm. Under conditions of calm, and little fresh-water inflow, turbidity dropped as low as 10 to 20 ppm in high salinity regions. Probably as many as half of all records showed turbidities in the range of 20 to 50 ppm and about 25 percent more in the range from 50 to 100 ppm.

Turbidity readings at a station did not always reflect turbidities over all of a bay, and demarkation lines between areas of high and low turbidity were often abrupt. On one occasion, turbidities ranging from 18 to 198 ppm were recorded with an electrophotometer while drifting a distance of less than 200 feet. In this case the boat was

drifted by the wind across a tidal tongue of high-salinity water which entered a small bay at a comparatively narrow pass. Such tongues of high-salinity water appear and are completely erased with great rapidity.

Even when a bay appears equally turbid everywhere, considerable local differences in turbidity measurements may occur; these are probably associated with small depth differences. In one test, three stations about one-half mile apart and forming a rough equilateral triangle showed turbidities (mean of three readings at each station) of 77 ppm, 32 ppm, and 63 ppm. Such differences are found in open water on a windy day; near exposed shores they may occur at nearly any time there is wind. Rolling of waves against a muddy shore often produces very local turbidities of 300 to 500 ppm. Those shorelines most exposed to wind action are favorite places for oyster leases because loose silt is kept scoured away.

After attaining a peak of high turbidity, the water usually clears quickly. For example, Hewatt's data show that at Station 51 in Barataria Bay on June 9, 1948, the turbidity reached 600 ppm at 1900 and three hours later had dropped to 200 ppm. The highest natural turbidity recorded by Hewatt was about 890 ppm at Station 51 on December 30, 1948, at about 1830. This dropped to 200 ppm in 2½ hours. Further clearing below 200 ppm is apt to be much slower for obvious reasons. Under conditions of continuous wind and heavy rainfall, turbidities may be maintained at a level of as much as 200 ppm over a period as long as 36 hours, and there is one record (at Station 51 on November 29, 1948) of turbidity remaining in excess of 300 ppm for a 24-hour period. This station showed turbidity levels fluctuating between 100 and 800 ppm from about noon of November 28 to midnight of November 30; during this period there were nine maxima in the range of 500 to 800 ppm and 10 corresponding minima between 100 and 400 ppm. High turbidities in the ranges indicated were never sustained longer than 2½ days. On the other hand, turbidities of around 100 ppm were often sustained for a week or more.

In the period from January to April turbidities are much higher than in other months of the year. From June to October turbidities are generally low. Other things being equal, high salinity areas have lower turbidities than do the low salinity regions, because of the precipitation effect of saline waters.

Almost from the beginning it was observed that mortality of oysters was in inverse proportion to the turbidity of the water. Stations with low salinity and high turbidity, such as Chene Fleur, had markedly lower mortality rates than did stations such as Sugar House in the southern part of Barataria. Mortality rates at this latter station were as high as any encountered anywhere. There the bottom was a hard mud-sand mixture with no loose silt, and it was frequently observed that the water would be comparatively clear there when it was turbid at other stations. Most commercial oyster leases are located along the margins of the bay, close to shores where wave action stirs up silt, and therefore have higher turbidities over them than can be found in open waters far from shore. Seasonal mortality rates were much higher in summer months when turbidity was generally low, and those months with frequent periods of high turbidity, January to April, had the lowest mortality rates of all. Because of these observations there was an early loss of interest in turbidity as a possible factor in mortality of oysters.

However, some experimental work has been done on the effects of turbidity. Methods were developed for maintaining constant high turbidity levels in aquaria, and a series of experiments was conducted to test the effect of silt suspensions on survival of oysters

(Mackin, 1956b). The data from these studies are presented in Table 8. Maximum turbidity used in experiments was 700 ppm. As shown in Table 8 this concentration produced no significant mortality in the experimental oysters. For details of the apparatus used see Mackin (1956b).

TABLE 8

Mortalities of oysters in experimental and control aquaria of the turbidity experiments. There were 20 oysters in all control and experimental tanks. x = experimental tank; c = control tank. All studies lasted 21 days

Experiment number	Maximum and minimum turbidity readings ppm	Approximate average turbidity ppm	Number oysters dying	Percent oysters dying	Dates
1-x	99–110	100	0	0.0	2–28–55 to 3–21–55
1-c	0–27	15	1	5.0	same
2-x	185–216	200	3	15.0	4– 5–55 to 4–26–55
3-x	290–318	300	2	10.0	same
2-3-c	4–30	15	4	20.0	same
4-x	390–420	400	7	35.0	4–27–55 to 5–18–55
5-x	490–532	500	4	20.0	same
4-5-c	0–27	10	5	25.0	same
6-x	495–720	590	3	15.0	5–23–55 to 6–12–55
7-x	639–765	710	4	20.0	same
6-7-c	0–45	15	3	15.0	same

WATER TEMPERATURE

Water temperature proved to be of primary importance. Mortalities of oysters were obviously correlated with temperature, since they were seasonal. The pattern of temperature change varies little from year to year. The graph, Fig. 14, shows temperature changes from July, 1948, to August, 1949. Lowest water temperature on record was 2°C in Bayou Rigaud on February 3, 1951. On that date air temperature dropped to —5°C, water in the margin of Bayou Rigaud had a thin film of ice, and many fish (mostly *Cynoscion nebulosus*) died as a direct or indirect result of the sudden cold. The highest water temperature recorded in open water was 35°C on the third of July, 1948, in Barataria Bay. Small bayous which drain from exposed mud flats may reach temperatures up to 40°C in summer. Most summer water temperatures range between 27° and 32°C.

Occasional very warm periods in winter have been observed. In January and February of 1957 there was a period in which water temperature was above 20°C for sixteen consecutive days (Mackin and Sparks, 1958).

SALINITY

Salinity in estuaries of the Mississippi Delta is probably the most variable component of the environment, and is dependent on a greater variety of modifying conditions than any other with which Project 9 had to deal. Much time and effort went into measurement of salinities.

Early references in the literature (Hewatt, 1951) consisted of "spot" records which

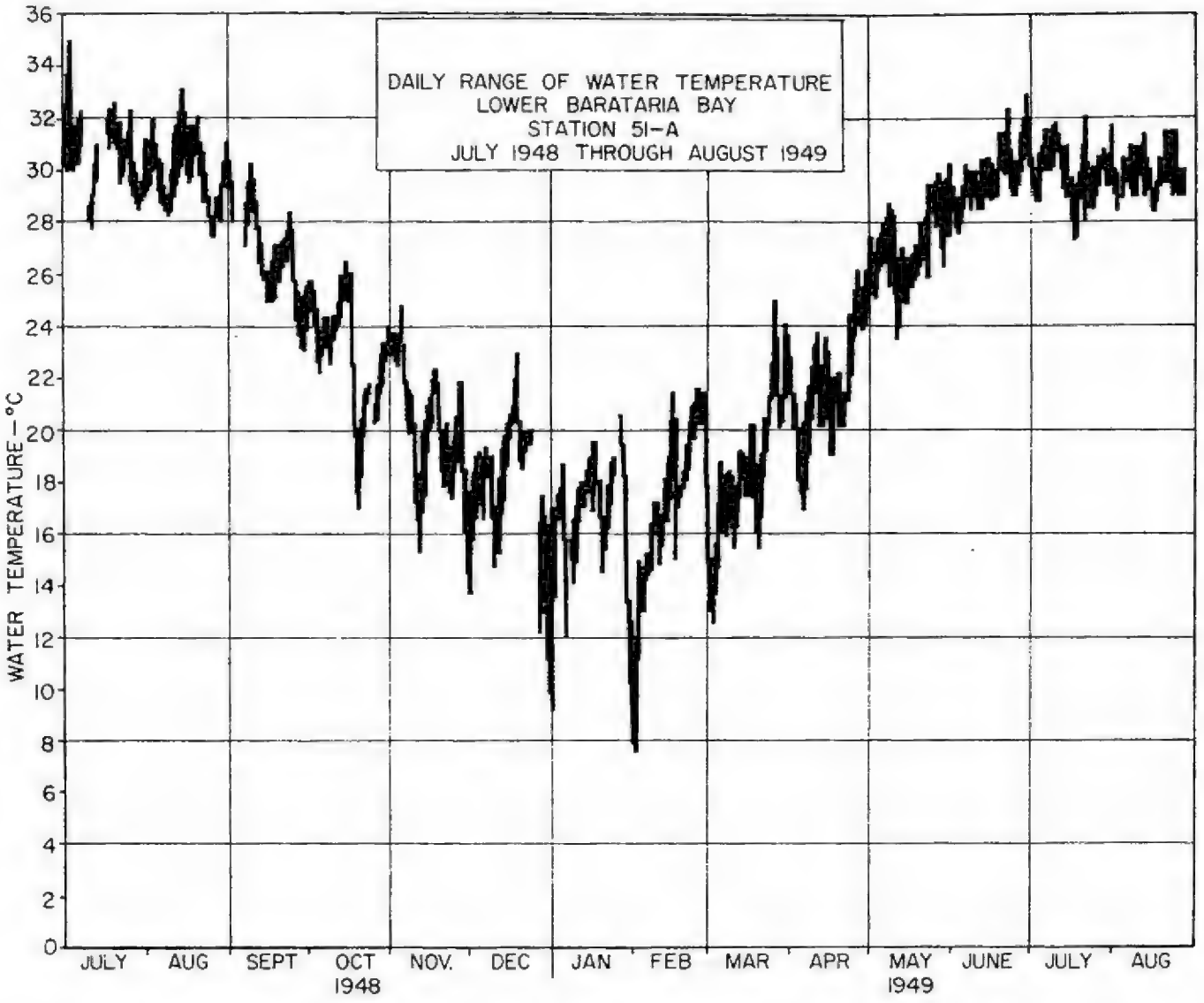

FIG. 14. Daily range of water temperature, lower Barataria Bay station 51-A, July 1948 through August 1949.

were often more confusing than they were enlightening; the measurements were too few, in some cases only means were presented without range, and correlative tidal conditions were usually not recorded.

The responsibility for salinity measurement in Project 9 fell to Dr. W. G. Hewatt. He carefully collected all available data from previous studies, and added many more data in the period from 1947 to 1950. The greater part of Hewatt's data were from the Barataria Bay region, but he also made extensive studies in bays to the west of Bayou Lafourche and in waterways on the lower west flank of the Mississippi River. These data are summarized by Hewatt (1951) in a single volume, but other extensive reports were issued in earlier years of the Project 9 studies (Hewatt, 1949).

Most of the salinity data were taken by means of instruments which measured the resistivity or conductivity of a column of water between two electrodes. Esterline Angus recorders activated by an electronic device were used (Jensen, 1948a). Temperature readings were taken from the column of water simultaneously with the resistivity readings. Calibrations were made against standard chloride titrations. There were 1635 checks for accuracy. The instruments proved to be accurate enough for the purpose intended, getting results in which the values recorded were within 1 ppt of the chemically measured value. The checks showed that 97.13 percent of the readings met with this requirement and 99.85 percent were within 1.5 ppt. The inaccuracies which occurred were mostly in the upper range of salinity. For a complete analysis of the calibrations

and accuracy checks, and details of the development of the instruments, see Parrack and Jensen (1950).

Eight instruments were constructed and used in the studies. One of these, used only briefly in Louisiana, was later installed in the Institute of Marine Science at Port Aransas, Texas. The other instruments were used in Barataria Bay, in the Buras region of western Placquemines Parish, and in areas to the west of Bayou Lafourche.

Two instruments were mounted on large speedboats and were used for daily mapping of isohalines of Barataria Bay and contiguous waters. The boats made "runs" along lines between set stations, which were so planned that daily salinity maps could be constructed. These runs began on April 11, 1947, and the last map was made on February 7, 1948. There were interruptions caused by storms and mechanical failures. For details see Hewatt (1951). From June, 1947, to June, 1948, a year's study, two boats made runs totaling about 100 miles daily. The isohaline charts were of very great value in the study of salinity gradients and the effects of rainfall, wind, and tides. The moving stations were used occasionally for study of outlying regions other than the Barataria basin.

Four fixed stations equipped with conductivity instruments were established in Barataria Bay: (1) in Bay Chene Fleur, where much field experimentation with oysters was carried out, (2) near the mouth of Bayou St. Denis, where much of the fresh water flowed into the bay, (3) at the oyster field experimental station in Bassa Bassa Bay, which is on the "conch line," and (4) in mid-lower Barataria Bay where the effects of Gulf water movements through Barataria Pass could be measured. Locations of these stations are shown in Fig. 15. See also Hewatt (1951), for a more complete description

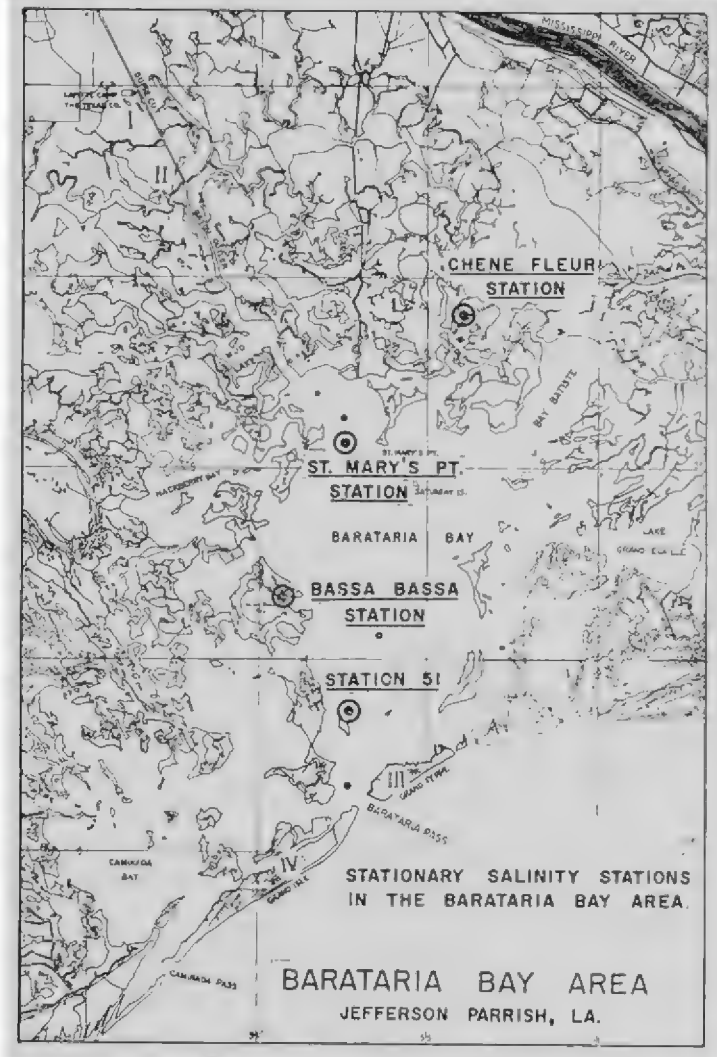

FIG. 15. Stationary salinity stations in the Barataria Bay area. 1948–1949.

of the stations and for exact periods of operation at the Barataria stations.

In the Terrebonne-Timbalier Bay areas salinity measurements were made with resistivity instruments: (1) at Bay Ste. Elaine, where a field laboratory for oyster studies was maintained, (2) in the Lake Barre oil field of the Texas Company, and (3) in Bayou Bas Bleu, above Lake Barre, at the location of another of the field experiment stations.

Supplementary to the measurements of salinity made by the instruments, many titrations were made. Regular stations were established (Hewatt, 1951) in the area east of Grand Bayou, on the west flank of the lower Delta. Results of these measurements were reported by Hewatt (1951).

It is not the intention here to present in detail the mass of salinity data collected by Hewatt and associates. Rather, it is hoped to use these data in an analysis of the relation of cyclic salinity changes to those factors regulating daily, seasonal, yearly, and multi-year cycles.

Salinity of Barataria Bay and its Relation to Rainfall

Hewatt (1955) collected and presented data on rainfall in and near the Barataria basin area, from the records of eleven New Orleans stations and four outlying stations of the U. S. Weather Bureau. These data show that:

(1) Over a period of thirty years ending in 1949, the mean annual precipitation was 62.10 inches.

(2) There were large fluctuations from the mean, especially in 1924 when only 40.73 inches fell and in 1929 when the total was 81.20 inches.

(3) When rainfall was above normal, the excess resulted from concentrated heavy rainfall in quite short periods rather than from small excesses distributed over the entire year. For example, in 1948, the excess was 11.98 inches for the year, but March alone had an excess of 12.11 inches and most of this fell in an eight-day period, one day of which had seven inches. Complete records of the effect of this concentrated deluge were made by the Research Foundation and the effect on oysters at different stations was observed. These data will be more fully analyzed below.

(4) On the other hand, when rainfall was below normal, the deficit resulted from generally small deficits distributed over more than half of the months of the year. The year 1931 is the best example. The deficit for that year was 9.57 inches. Nine months of the year were below normal and the remaining three averaged only 1.91 inches above normal. In 1931 and the following year, 1932, extensive mortalities of oysters occurred.

(5) Barlow (1955) has called attention to the fact that in a forty-year period there were 316 out of 480 months with rainfall below the mean, and that there were numerous periods of two months or more of deficit. He presented these data as follows (Barlow's analysis was based on Hewatt's data):

Number of periods of less than mean rainfall, the duration of which was	
2 months	20
3 months	12
4 months	12
5 months	4
6 months	7

7 months	7
8 months	4
9 months	2

Barlow pointed out that, according to Hewatt's data, a two-month period of rainfall deficit would result in a measurable rise in salinity in Barataria, whereas a shorter period had little apparent effect. Exceptionally heavy rains, on the other hand, may produce a precipitous decrease in salinity in a few days time, an effect which may last for a month or more without added rainfall above normal. After a drouth period normal rains produce little effect on salinity, probably due to the capacity of low lying marsh to absorb rainfall and return it by transpiration to the atmosphere.

Marmer (1948) showed that the excess of ebb over flood in Barataria, Abel, Caminada, and Quatre Bayou passes was 430 million cubic feet, which he considered to be the amount of fresh water draining into Greater Barataria Bay daily during the period of his study, August 12 to 28, 1947. Since June and July of 1947 both showed rainfall deficits and August, 1947, was only slightly over normal (0.8 inch), the period of his study was either about normal or less than normal for rainfall, and the average daily fresh-water inflow is probably close to or above the figure given by Marmer. Barataria Pass carried most of the net out-flow (209 million cubic feet). The total of fresh water (or equivalent) volume leaving the bay at all passes (430 million cubic feet) was about 8.0 percent of the total ebb volume (5308 million cubic feet).

Jensen (1948a) gives the area of the Barataria Bay watershed as 1914.65 square miles. Based on this area, calculations show that the mean daily rainfall in the area would be about 1 billion cubic feet. Thus the mean return to the atmosphere by transpiration and evaporation would be about 57 percent of the rainfall. It must be much less than this following periods of excessively heavy rainfall, but in periods of long continued subnormal rainfall in summer when temperatures are high and vegetation growth is lush, the return to the atmosphere may be even greater.

In March, 1949, the effect on salinity of a general heavy rainfall concentrated in a short period of time was recorded by Hewatt (1951). During the period from the third to the tenth of March 15.85 inches of rain fell in the Barataria watershed. The effect on the salinity at Bay Chene Fleur above Barataria and near the mouth of Bayou St. Denis, the major fresh-water entry into the Bay, was measurable on the second day. By the tenth of March the salinity at these stations had been depressed from above 20 ppt to below 3 ppt. The Chene Fleur station did not regain a salinity as high as 10 ppt until June, nearly three months later. This station was too far inland to be materially affected by influx of flood tide waters. Far down bay at Station 51 the rains had less effect, but still depressed salinities from a range between 20 ppt and 30 ppt to a range between about 5 ppt and 20 ppt. At this station the wedge of high-salinity water pushing in from the Gulf caused daily saltations from about 5 to 20 ppt during the middle part of March, and the effect of the rains was largely nullified by the end of March.

The months of April, May, and June following the torrential rains described above were 2.4 to 3.1 inches below normal in rainfall; the latter part of March, following cessation of the rains, was also below normal. The continued depression of salinity at the Chene Fleur Station therefore was due to continued storage of some of the rain water in the marshes, whence it fed more and more slowly into Barataria Bay. Resistance of the fresh water to mixture with high-salinity water from the Gulf also had some effect. There

was a sharp gradient at the interface between high-salinity and low-salinity water masses. This is shown clearly by the hourly graphs of salinity for Station 51 in lower Barataria Bay (Fig. 16, from Hewatt, 1951). According to this figure the maximum effect of the rains was felt on March 12, when so much fresh water had penetrated the lower bay that the flood tide wedge of high-salinity water failed to reach Station 51.

Salinities in Barataria Bay after Several Months of Rainfall Deficit

The effect of cumulative rainfall deficit is exemplified by the salinity records for the months of June to October, 1947. This period is especially interesting because from the end of June to September mean tidal level normally becomes higher by approximately 5 inches (Marmer, 1954), and the highest mean monthly tide level of the year occurs in September.

Hewatt (1951) has recorded the rainfall during the period under study. His records show that the deficit was 0.24 inch in June, 1947, and 3.68 inches in July; August recorded a small excess of 0.8 inch and September had a deficit of 1.60 inches. Rainfall for the general area and salinity at five stations in Barataria Bay are shown in Fig. 17. The data are taken from Hewatt (1949). These stations are arranged from the upper

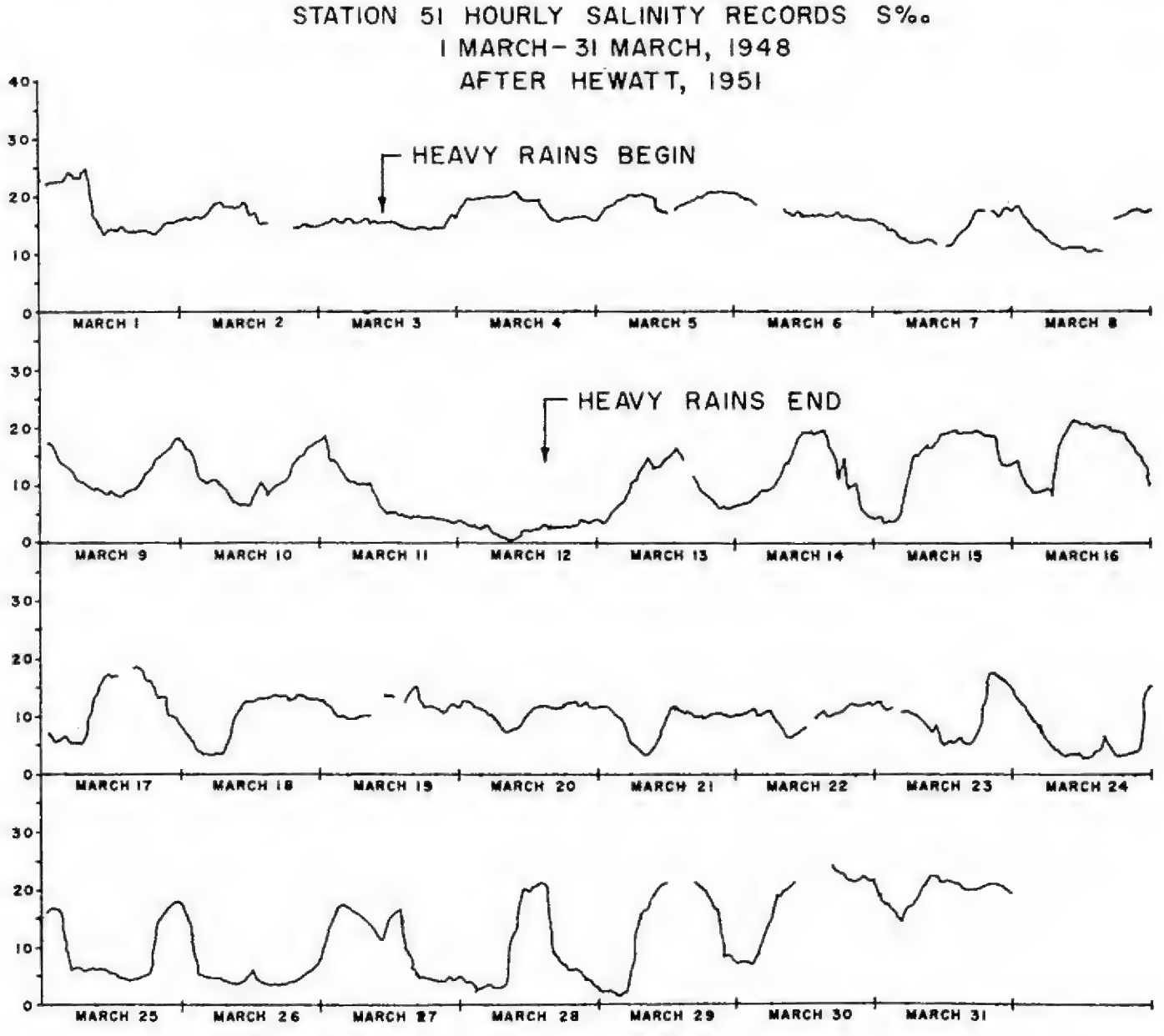

Fig. 16. Station 51, hourly salinity records S 0/00, 1 March–31 March, 1948, after Hewatt, 1951. The effect of heavy rains on salinity in lower Barataria Bay is shown.

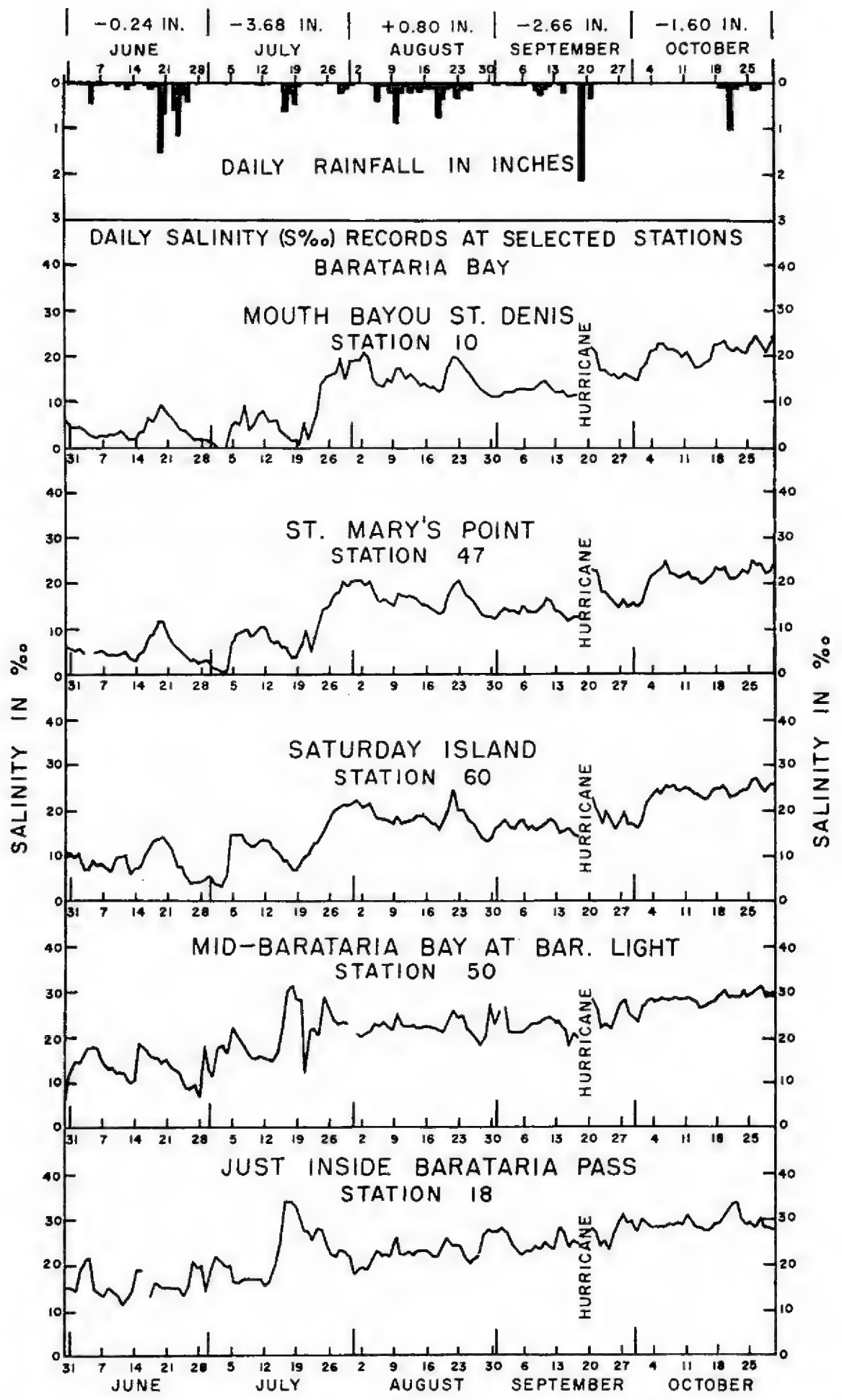

FIG. 17. Salinity changes in summer and fall at five stations in Barataria Bay, La., 1947. Rainfall scale is included. The effect of continued sub-normal rainfall over several months is shown.

bay (generally low salinity) through graded steps to the south, ending with Station 18 which is situated just inside the bay at Barataria Pass.

The salinities show a gradual increase during the "drouth" period. Note that it is not a drouth in the usual sense, for rain was recorded on 21 days in August. At

Station 10, where salinity is normally quite low, it ranged between zero and 10 ppt in June, increased to the end of October, and ended with the last 10 days fluctuating between 21 ppt and 25 ppt. At the other extreme, Station 18, just inside Barataria Pass, started in June with salinity fluctuating between 11 ppt and 22 ppt, and ended in October with a range between 27 ppt and 34 ppt. Stations 50 and 18 are in "normal" conch territory; the three northerly stations do not ordinarily have salinity high enough to permit these predators to live. All of the stations from late July to the end of October attained salinities well within the tolerance of *Thais haemastoma.* Those stations which have one or more annual low salinity periods, long enough to kill conchs, are the ones free from conchs under normal conditions. The mean salinity at such stations may be well above the tolerance limit; it is the extremes, not the means, that limit the depredations of *Thais.*

The increasing salinity in the case discussed here probably was enhanced considerably by the increasing level of the tide. Water levels in the marshes above Barataria Bay must increase equally with Gulf levels to offset the effect of higher mean tide. The latter would act as a sort of a dam, which is shifted farther inland in the case of rainfall deficit of long duration. Thus the rising salinity was produced not only by deficit of rainfall; there was also a decrease in the proportion of fresh water to sea water in the bay because of an increase in volume of sea water.

In the winter months this effect is reversed. From September or October, when highest mean tide level is reached, there is a rapid lowering of mean tide level until January. In this period of three months, mean tide level drops about 0.9 foot (nearly 11 inches). This lowering of the "salt-water dam" increases the effect of rainfall in lowering the salinity. For example, the period of rainfall deficit shown in Fig. 17 was followed by excess rainfall of nearly 12 inches during November and early December, 1947. This was sufficient to keep salinity levels low in the upper bay during the following two-month period of deficit rainfall, until February, 1948. Stations in the lower bay were definitely depressed, but recovered in a period of less than one month.

Effect of the Mississippi River

The Mississippi River once exercised control over salinity in all waters of the Delta marshland. With the construction of levees this major influence was steadily reduced (Gunter, 1952). Even after levees were complete and about as they are today, frequent crevasses produced highly effective flushing of major portions of the Delta. The last of these, so far as the west Delta flank is concerned, occurred in 1927. Since that time the Mississippi River has had some effect in lowering salinities in Barataria Bay but only when the river is in flood stage. Fresh water discharged into the Gulf through Southwest Pass and other mouths of the river is stratified over the highly saline Gulf water, and is carried into Barataria and other passes by tidal movement. This may be very effective in lowering bay salinities during periods of high river discharge, especially from Sandy Point to Bastian Bay on the lower west Delta flank. A lesser effect is sometimes felt in Barataria Bay. The surface currents from the end of the Delta pass westward along the coast and are progressively closer inshore from winter to summer. The fresh water layer may get deep enough to block the deeper saline water from the passes. The effect is transient.

The effect of encroachment of low-salinity water from the Gulf into Bastian Bay is

shown by records taken by Hewatt (1951) on March 23, 1948, and April 24, 1948. The river was in high stage in March and April of that year (128.4 and 120.3 percent of the means respectively for those months). For comparison, data from December 23, 1947, are given. The data are presented in Table 9.

TABLE 9

Effect of Mississippi River level on salinity in Bastian Bay. Salinity measured by titrating spot samples of water

Station	Salinity Dec. 23, 1947; river level 78% of mean; ppt	Salinity Mar. 23, 1948; river level 128% of mean; ppt	Salinity April 24, 1948; river level 120% of mean; ppt
Bayou Cook above Bastian Bay	18.4	13.2	12.9
Upper Bastian Bay	23.5	12.7	13.0
Lower Bastian Bay	26.0	9.8	11.8
Pass to Gulf	31.3	6.5	11.5

The greater effect on March 23 may be due to lower Gulf level at that date, and possibly difference in tide stage. With the river level at only 78 percent of the mean on December 23, 1947, it is obvious that the fresh water dilution of Bastian Bay came from land drainage, which is small at this point on the Delta flank. The small bays of the lower west Delta flank are dependent on Mississippi flood conditions for most of their fresh water. It is for this reason that oyster mortalities associated with high salinity have been frequent throughout the recorded history of the area.

Two figures from Hewatt (1951) are reproduced to present ranges and modes of salinity (Figs. 18 and 19), at two stations. Station 17 is in a small bay (Chene Fleur) above Barataria Bay proper and is in the low-salinity zone of oyster production. Station 51 is in the middle of lower Barataria Bay. The difference between the ranges at the two stations is less than the daily tidal variation at one station in the high-salinity area. The modes, however, are significantly different. The low-salinity station at Chene Fleur tends to have two modes, one produced by low salinities in March, April, and May, and another by relatively high salinities in July, August, and September. Other months tend to have salinities spread more or less equally from low to high. It is of interest that the high-salinity months are the months of mean high precipitation. Rising mean tide levels, transpiration from vegetation, and evaporation more than offset the higher rainfall. High precipitation during the summer is succeeded by several months of mean low rainfall. During the fall months, salinity levels usually show a gradual increase and high-salinity waters move into the upper reaches of the bays and bayous above Barataria Bay proper.

SALINITY GRADIENTS FROM EAST TO WEST IN BARATARIA BAY

So far the discussion has involved only salinity gradients from the passes on the south to the north. There is, however, a very definite east-west gradient also. Most fresh water drains into Barataria Bay from the northwest, through Bayou St. Denis, Grand Bayou, Bayou Cholas, and Wilkinson Canal. Wilkinson Bayou and other northeast entry ways are less important. There is a relatively small watershed to the east, bounded by the Mississippi River levee. The freshwater inflow tends to move down the west side of the

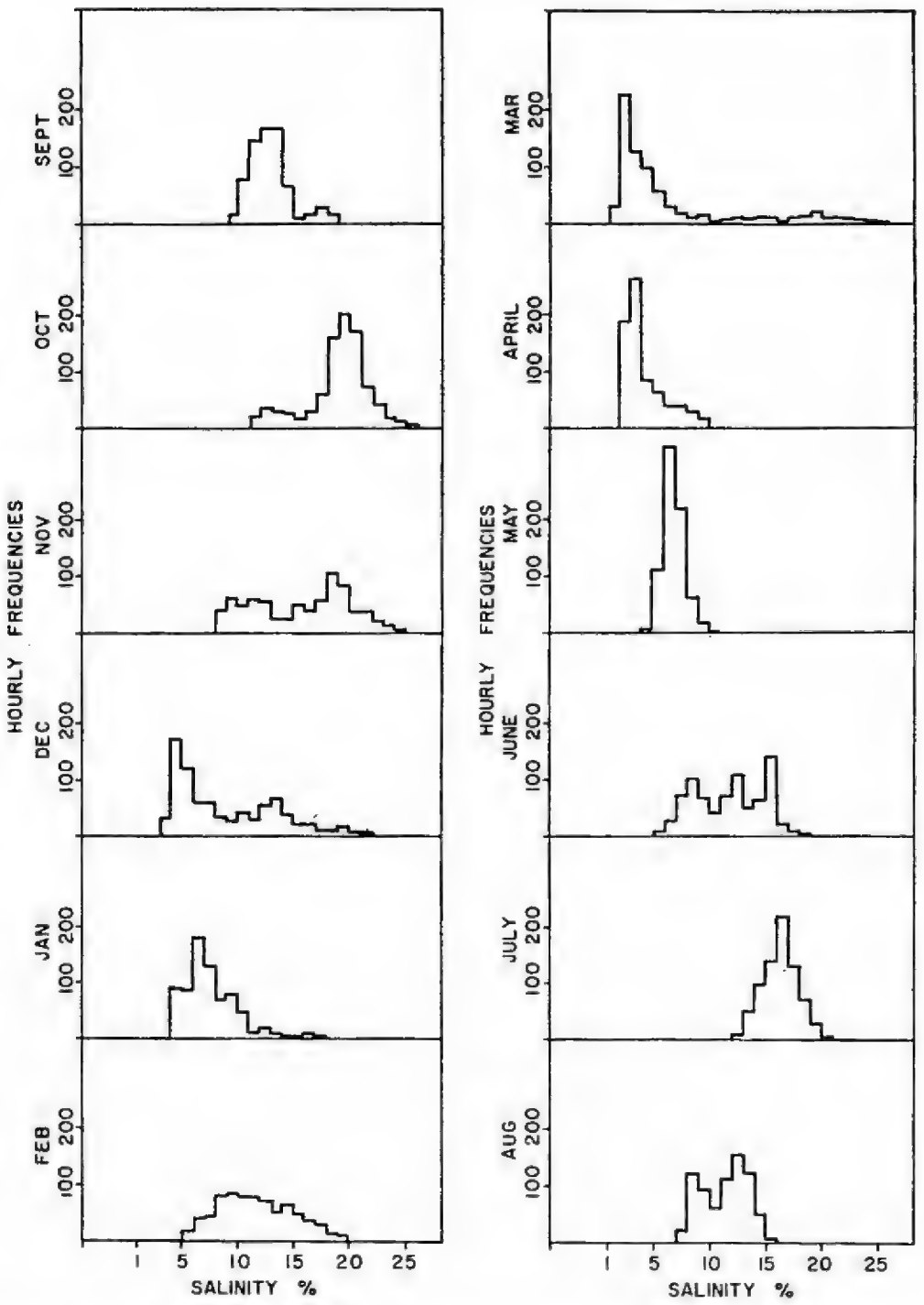

FIG. 18. Salinity frequencies at station 17, Chene Fleur Bay, September, 1957, to August, 1958, after Hewatt, 1951.

bay, aided by Coriolis force and prevailing easterly winds. These conditions often produce isohalines which traverse the bay from south to north. Pockets of high salinity waters in the east and northeast parts of the bay are of common occurrence (Fig. 20). Tributary bays, such as Lake Grande Ecaille, lying to the east of Barataria Bay proper, often show comparatively little effect of freshets. More nearly stable high salinity is therefore characteristic of the easterly areas.

RISING SALINITY DURING THE FIRST HALF OF THE 20TH CENTURY

Many references in the literature indicate that the salinity level in the oyster-producing bays of Louisiana has been increasing since the beginning of the century. In fact, the beginning of this phenomenon probably far antedates 1900.

Early salinity records only doubtfully support this thesis. A careful study of the early records (Hewatt, 1951) leaves the impression that it could be true. But the spotty nature

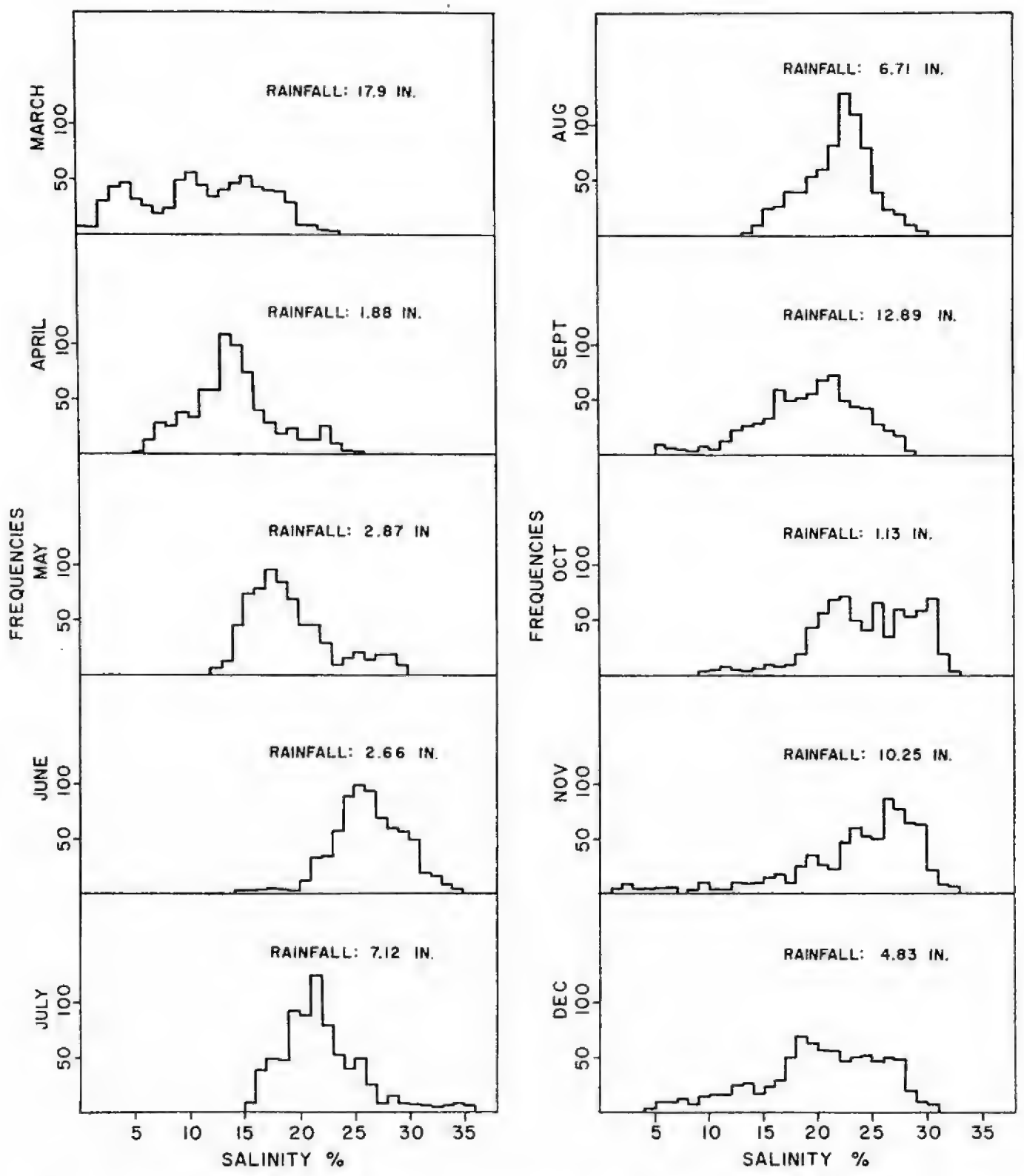

FIG. 19. Salinity frequencies at station 51, Barataria Bay, Louisiana, from March to December 1948, after Hewatt, 1951.

of these records makes a valid comparison impossible. They prove only that in particular areas there were periods of both high and low salinities, with no indication that one or the other predominated.

However, the references in the literature indicate that, at about the beginning of the century, generally lower salinities prevailed. To review some of these briefly, Cary (1906) stated that freshets were "by far the most formidable destructive agent to be encountered" by oyster culturists. Again (1907a, 1907b) he repeated his earlier statement, but said that this danger was less in Terrebonne Parish than was the case farther west. Gates (1910) warned against selection of leases for oyster culture in low-salinity areas. Moore (1899) stated that the probable reason why oysters had never been produced in the northern part of Barataria was because of the "low normal salinity,"

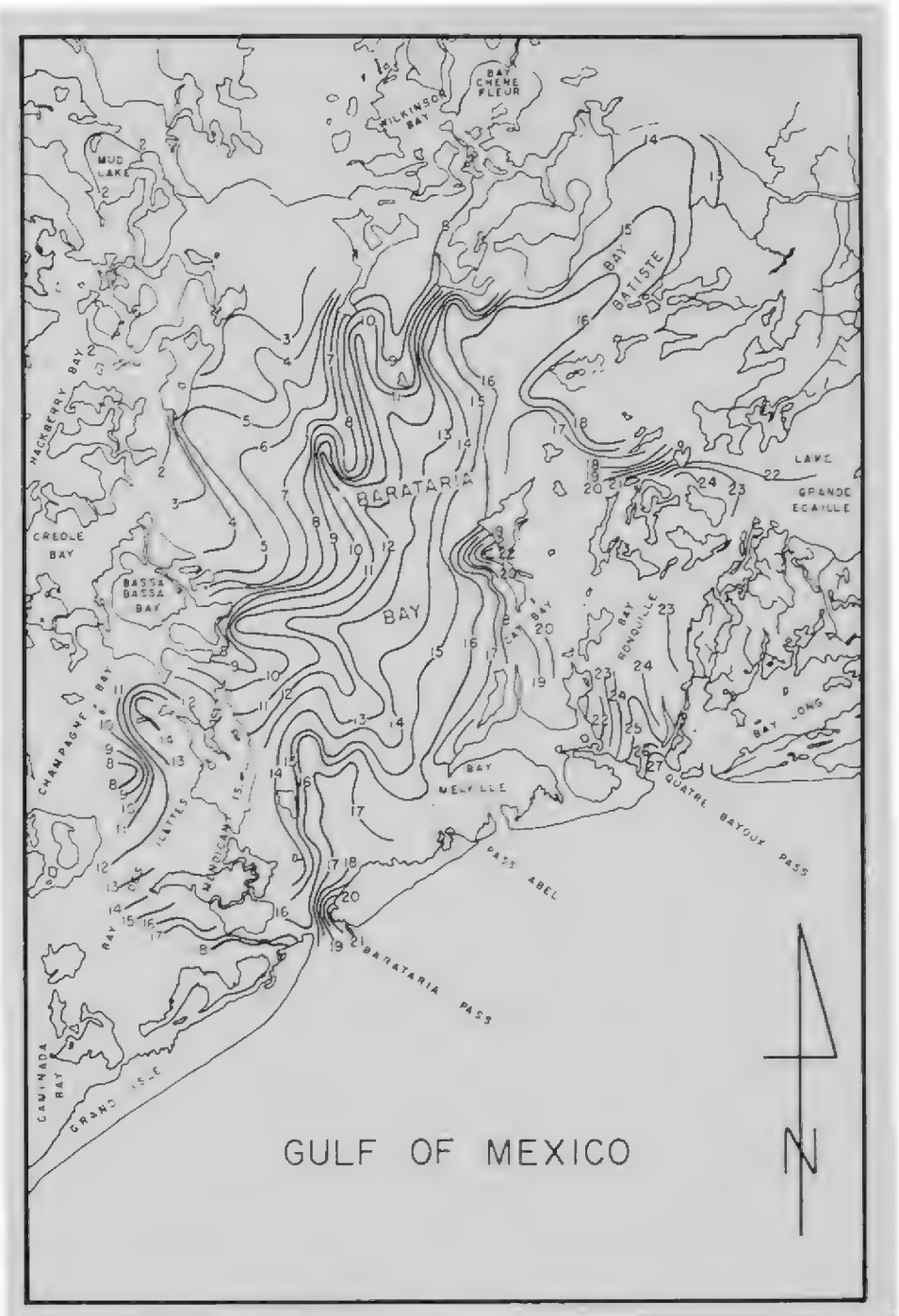

FIG. 20. Isohalines in Barataria Bay, March 10, 1949. The gradation of salinities from west to east is caused by large volumes of fresh water from excessive rainfall which fell in the Barataria watershed from March 3 to March 10, 1949. After Hewatt, 1951.

and reported that a dead reef in Little Lake (of Lafourche Parish) may have been destroyed by fresh water. He stated that Terrebonne Bay was too fresh for the conch (*Thais haemastoma*) to live. Other records indicate that this was not true of all of the bay, but it may have been true for the more northerly half. Baylor (1904) advocated construction of locks and dams for protection against freshets and crevasses, which he thought constituted the greatest danger to Louisiana oysters. The locks and dams referred to were on the Arkansas and Missouri Rivers. In 1908 the Oyster Commission of Louisiana recommended that the Baptiste Collete Gap on the lower east bank of the Mississippi River be closed to cut off fresh water from certain areas east of the river.

The opinions in this matter were not unanimous, even at the turn of the century. Apparently low salinity effects were observable in certain localities while in others the troubles were either indeterminate or associated with predation in high salinities. As the years passed, low salinity, except as associated with crevasses or spillway openings, ceased to be mentioned and the association of mortalities with encroachment of high salinity became more and more frequent. A detailed discussion of the references is

given in the section dealing with the oyster industry and will not be repeated here. By about 1940 to 1950 the concept of increasing salinity was firmly established in the literature.

Characteristic of the reports of increasing salinity was the absence of actual salinity data supporting the thesis. It was not clear whether or not the "increasing" salinity meant actual progression to a higher level, the maxima of which exceeded past maxima, or whether the number of recurrences of high salinity periods was increased. Perhaps it may have been an increase in duration of high salinity periods rather than increase in number of such periods.

An exception to the rule that reports of increasing salinity were not documented has recently appeared. St. Amant (1955) presented salinity data for Sister Lake covering the period from1950 to 1955 which show a rapid increase. It paralleled collapse of the area as an oyster production center. From the beginning of the century Sister Lake had been an important seed producing area.

In spite of the fact that most statements are not supported by actual data, it is believed that there has been a progressive change toward high salinities. It is the intention here to summarize the factors which are thought to have produced progressive change in salinities in Louisiana bays, with special reference to Barataria and its satellite bays and bayous. Some of the effects are well established while others are largely inferential.

(1) Levees. Probably the most important of the factors having to do with salinity rise was the construction of the extensive levee system of the Mississippi. Gunter (1952) has reviewed the history of the levees. Important milestones should be pointed out. By 1904 the system of levees cutting off river water from the delta basins between the Mississippi River and the Atchafalaya basin was complete and Bayou Lafourche was cut off from the river in that year. It was the last of the major distributaries to be so severed. But frequent crevasses continued to bring intermittent relief until 1927 when the levees were shattered by a vast flood. After that the U. S. Army Engineers assumed responsibility for maintenance and built the levees higher. They completed the system in 1935, when it included 2130 miles of confining embankments. The date of 1927 marked the end of the crevasses which let river water into the high-mortality region west of the river. Elimination of crevasses meant the loss of one of the most effective agencies controlling enemies and competitors of oysters.

(2) Rise in Gulf level. Marmer (1954) has shown that there has been a steady rise in Gulf level beginning early in the century. Rise in Gulf level means a lessened gradient from the watershed into the bays with consequent retention in the fresh-water marshes of a greater quantity of fresh water. At the same time subsidence has deepened the bays (see summary of references by Gunter, 1952). The increased depth and higher water level have increased the volume of water in the bays, requiring a larger fresh-water volume for dilution to any given salinity. Since it is apparent that the required fresh-water increase has not been provided, an increase in salinity necessarily took place.

(3) Blockage of drainage distributaries in the fresh-water marsh. Water hyacinths probably were introduced into Louisiana in 1884 (Wunderlich, 1941), and alligator weed appeared at about the same time. By 1900 these exotic plants had spread and increased until they were a serious menace to navigation. Much study and effort was put into attempts to control these pests, with considerable success in the larger, more important bayous. But in the minor distributaries, and in ponds and lakes of the fresh-water marsh, little has ever been done. Most of these minor distributaries and much of

the pond area in the marshes became completely filled with the plants, and alligator weed invaded the land areas of the marsh.

These two exotic plants contributed to the deficit of fresh water in the oyster-producing area in two ways. First, they prevented movement of water by mechanical blockage, and second, they returned large amounts of moisture to the air by transpiration. There is now a well-defined zone in the fresh-water marshes that is without drainage except for the major waterways, natural bayous and dredged canals, which are kept clear for navigation purposes. Vast areas of the fresh-water marshes are undrained because the bayous follow natural levees which are higher than the intervening marsh basins. Water tends to drain away from the bayous rather than into them. On the other hand, artificial canals cut through all levels, and drain marshes better than the natural bayous. There is no known way to measure the total effect of hyacinth and alligator weed, but they must have large effects.

(4) Disintegration of shorelines and islands. For many years the bays of the west Delta flank depression have been widening and deepening. Shorelines disintegrate at a fast rate, and whole islands have been observed to disappear within the last 10 years. Increased wind fetch and consequent increased scour has helped deepen the bays (Price, 1947). Sediments of the Mississippi River formerly deposited in the Delta are now deposited in the Gulf proper. Subsidence has contributed to the depth of the bays. These increased basins now contain much more water than was the case at the beginning of the century. The additional water comes from the Gulf, while the fresh water supply has decreased.

Doubtfully, there may be a decreased flow of the Mississippi River and a tendency to change the pattern of flow. This cannot be determined with certainty. If it is true, the small bays of the lower west Delta flank may get less fresh water, at critical periods, from this source.

There are certain factors that have not contributed to increased salinities. Rainfall has not decreased perceptibly in the last thirty years. According to the data assembled by Hewatt (1955), rainfall in the Barataria watershed was somewhat greater in the period of 1940 to 1949 than it was in the decade from 1920 to 1929. Neither has there been any detectable change in the pattern of rainfall.

Canal dredging, sometimes assigned a role in increasing salinities (McConnell, 1956), can hardly be a contributor. Additional canals do not admit more water from the Gulf, since the depth to which the bays fill on flood tide is controlled by Gulf level, not by the number of access waterways. The saline marshes are already well drained, not being affected by blockage as are the fresh-water marshes. Canals dredged in the fresh-water marsh and connecting with the saline bays might conceivably be of benefit in reducing salinities in the highly saline areas. If the fresh-water areas were completely leveed from saline waters, canals connecting the two areas would admit saline waters to the fresh-water areas and vice-versa. In such a hypothetical situation, the net effects of the canals would damage the fresh-water areas especially in rice-producing localities, but they would benefit the oyster-producing region by lowering the salinity, and possibly by bringing in more nutrient substances.

Biological Significance of Salinity

A drop of salinity below about 5 ppt, if long continued, will result in mortality of oysters. The time required to kill oysters varies according to the temperature and

season, the previous condition of the oysters, their age, and perhaps other factors. Because of their ability to keep their shells closed for long periods, oysters can survive short periods of low salinity, even fresh water, that will kill most of the other organisms that live on oyster beds. Near the inland limit of the oyster's range, the fauna of oyster beds is normally composed of relatively few species. Some of these species, that share with the oyster the ability to tolerate low salinity, become far more abundant in this marginal zone than in higher salinity, presumably because of the relative scarcity of their enemies. These are called "low-salinity species." Some of them, e.g. *Rangia cuneata*, *Congeria leucopheata*, and *Rhithropanopeus harrisii*, can tolerate much lower salinity than the oyster and reach their maximum abundance even farther inland, beyond the range of the oyster. Others, such as *Cliona truitti*, do not extend quite as far inland as the oyster because they are less tolerant of extremely low salinities.

Increased salinity, unlike fresh water, does not have an adverse effect on the oyster directly. A continued high salinity, in southern waters, is unfavorable to oysters because it is favorable to their enemies: shell-boring organisms like boring sponges (*Cliona spp.*, especially *C. celata*), the boring clam *Martesia smithii*, and the mudworm *Polydora websteri*; competitors for food and space like sea squirts (tunicates) and slipper limpets (*Crepidula spp.*); predators of spat and older oysters like *Thais haemastoma* and *Menippe mercenaria*; and disease-producing parasites such as *Dermocystidium marinum*. An increase in salinity large enough and long enough to let these enemies become established will result in an increased rate of mortality among the oysters below the low tide level. In southern waters of continuously high salinity, oysters are confined almost entirely to the intertidal zone. Areas with normally high salinity become much more favorable to the development and maintenance of large populations of oysters after a freshet wipes out most of the high-salinity fauna.

The following lists name some of the commoner species found on Louisiana oyster beds. "High-salinity" species are those that become more abundant as salinity increases, up to 35 ppt. As you go up the bay, or as salinity decreases, these species drop out one by one until only those most tolerant of low salinity remain. These remaining species are those named in the second list, "species more or less independent of salinity." Species that not only tolerate low salinity, but become more abundant as salinity nears the lowest level that oysters can stand, are named in the third list, "low-salinity species."

Salinity Relations of Some Common Animals on Louisiana Oyster Beds

1. High-salinity species:

Cliona celata Grant, sulphur sponge, boring sponge
Haliclona sp., encrusting sponge
Astrangia astreiformis Milne Edwards and Haime, solitary coral
Mercenaria sp. clam
Martesia smithii Tryon, shell-boring clam
Corambella baratariae Harry, nudibranch, sea slug
Crepidula plana Say, flat slipper limpet
Thais haemastoma haysae Clench, conch, oyster drill
Anachis obesa (Adams), fat dove shell
Anachis avara Say, greedy dove shell
Nassarius vibex (Say), mud snail
Eupomatus dianthus (Verrill), calcareous tube worm

Clibanarius vittatus (Bosc), hermit crab
Crangon armillatus (Milne Edwards), *C. heterochaelis* (Say), pistol shrimps
Menippe mercenaria (Say), stone crab
Callinectes danaë Smith, little blue crab
Molgula manhattensis (Dekay), sea squirt

2. Species more or less independent of salinity:

Cliona truitti Old, small boring sponge
Sea anemones (including *Aiptasiomorpha texaensis* Carlgren and Hedgpeth)
Stylochus ellipticus (Girard), flatworm
Bugula neritina (Linn.), bryozoan (species not certain)
Membranipora sp., encrusting bryzoan
Polydora websteri Hartman, mudworm
Spiochaetopterus oculatus Webster, tube worm
Nereids (probably including *Nereis pelagica occidentalis* Hartman and *Neanthes succinea* (Frey and Leuckart))
Odostomia impressa Say, parasitic snail
Balanus spp. (including *B. eburneus* Gould and probably *B. improvisus* Darwin), barnacle
Palaemonetes spp. (including *P. vulgaris* and *P. pugio*, certainly, and possibly *P. intermedius*), grass shrimp
Unidentified gammarid amphipods
Corophium lacustre Vanhöffen, *C. louisianum* Shoemaker, mud tube amphipods
Callinectes sapidus Rathbun, blue crab
Eurypanopeus depressus (Smith), mud crab
Panopeus herbstii Milne Edwards, mud crab
Opsanus beta (Goode and Bean), oyster dog, toadfish
Gobiesox strumosus Cope, sucker fish
Gobiosoma bosci (Lacépède), goby
Hypsoblennius ionthas (Jordan and Gilbert), blenny

3. Low-salinity species:

Brachidontes recurvus (Rafinesque), hooked mussel
Congeria leucophaeata Conrad, river mussel
Rangia cuneata Gray, marsh clam
Neritina reclivata Say, brackish water snail

Hydrocarbons in Muds and Water

Very early in the investigation, Texas A & M Research Foundation chemists began trying to determine what muds and waters of the Louisiana oyster-producing area contained hydrocarbons of petroleum origin, and how much. The aim of these studies was to find out whether or not oysters were subjected to oil field pollution, as claimed, and if so, to what areas the pollution extended, and whether or not there was a correlation between amounts of pollution and mortality of oysters. A knowledge of concentrations of petroleum and bleedwater occurring in the bays was necessary also for interpretation of the results of laboratory and field experiments on oysters.

Gowanloch (1936) speculated that oil was carried to the bottom of bays and bayous, and there liberated highly toxic substances. He made no attempt to determine the concentrations of petroleum or whether there was pollution on the beds where oysters died. In fact, Gowanloch (1936, p. 239) noted that there was no relation between the locations of oil developments and those of oyster beds where oysters died. Smith, in Galtsoff *et al.* (1935), stated that, "oil pollution will last over a long period, for the oil is carried to the bottom by suspended mud particles and released from time to time by storms, tonging, or dredging." However, Smith also failed to analyze muds of the bays to determine whether oil had actually been carried to the bottom of oyster beds or elsewhere, or how long was its life if carried to the bottom. He observed slicks at Lake Barre oil field and also found oil in a canal connected with Bayou Lafourche, two widely separated areas. He made no mention of seeing oil at 423 stations occupied by him for taking hydrographic data.

During 1946, the Division of Oysters and Water Bottoms of the Louisiana Department of Wildlife and Fisheries instituted a mud sampling program aimed at determining the hydrocarbon content of muds in oil leases and in oyster beds. These samples were taken by E. F. Lindsey and F. H. Webster, who reported the locations of samples to Director James N. McConnell of the Division of Oysters and Water Bottoms. Owen (1955) stated that results of these determinations were contained in Appendix 17 of his report, but our copy of this report has no such appendix. Owen remarked, "It appears that the selection of these samples was not completely random, and that little or no effort was made to get a comparative picture of the amount of ether-soluble materials in the muds. The paucity of data from muds collected in areas away from oil wells is apparent."

In his own study of extractable materials from muds, Owen compared areas of low mortality (mortality index 30.0) and areas of high mortality ("mortality index" 75.0). Owen's "index" was apparently the percentage mortality. His data show that there was no difference in the concentrations of ether-soluble substances in muds in the two areas. Owen took a total of 83 samples, of which 21 were in his low mortality area and 62 in the high mortality area (Bastian Bay—Barataria Bay). The average value in all extractions (by two methods) was 0.0216 percent in the low mortality area, and 0.0211 percent in the high mortality area. Owen considered these to be equivalent values.

Dr. Fred W. Jensen and Dr. William M. Potts of the Chemistry Department of Texas A & M College made extensive studies of the "hydrocarbon" extractives of muds in Barataria Bay and adjacent waters. They also measured hydrocarbons in muds around bleedwater outlets at various oil fields and compared the amounts with those in oyster-producing areas. Some of these oil fields are in fresh-water or brackish marshes and bays, connected with the saline marshes by various waterways. Others are in the oyster-producing territory. Jensen and Potts presented their data in several small preliminary reports and in one final summarization (Jensen and Potts, 1953).

Jensen (1948b) also made efforts to determine, by means of infra-red spectroscopy, whether the extractives of muds were petroleum fractions, some other class of hydrocarbon compound, or possibly substances other than hydrocarbons. He suspected that some of the extractives originated in marsh plants and made certain studies aimed at testing that theory. (Because of a reasonable doubt that all substances extracted from natural muds were really hydrocarbons, the word "hydrocarbons" has been enclosed in quotation marks throughout this section, except where there was reason to believe that the bulk of this extracted material actually was hydrocarbons.)

In addition to this approach, Jensen (1950) collected data on total production of bleedwater in oil fields of the Barataria watershed. With these data, and figures developed by Marmer (1947, 1948), he calculated dilutions of bleedwater and crude petroleum in the bay waters. These were not intended to be accurate representations of actual amounts present, but the maximum possible amounts.

In addition to Drs. Jensen and Potts and their staff, the Horvitz Laboratories of Houston, Texas, was engaged to make determinations of more volatile fractions of crude petroleum in muds and water. No effort was made to determine the origin of these fractions. The Horvitz Laboratory makes a specialty of detecting and measuring minute amounts of hydrocarbons in soils and muds. All of the Horvitz Laboratory analyses were reported by Jensen.

Methods of Sampling

In the early days of these researches, samples of mud were taken by Ekman dredge. Comparatively few were taken in this manner, however. A corer of the Emory-Dietz type was soon substituted for the dredge and was used for all subsequent sampling. The samples consisted of short cores, 40 inches in length, more or less. It was quickly learned that a single core would not reflect accurately the conditions at a station. A system was worked out in which nine cores were taken at each station in a pattern consisting of one central core and two concentric rings of four cores each around the center. The entire pattern had a diameter of about 20 to 40 feet. Extractions were made separately on each of nine cores, and a composite of the cores was also extracted for comparison. When it became evident that the mean of the nine separate cores checked closely with the composite of all of them, only the composite was thereafter analyzed. In addition to the composite sample, the core at the center of the pattern was sampled at three levels, the top, middle, and bottom four inches. Extractions were made and the amounts of extracted material reported separately for each of the three sections.

Both the "wet" and "dry" methods of extraction of muds were used (Potts, 1949). Samples were fixed and shipped in sealed cans or jars, after being collected in the field by a specially organized crew of chemists and assistants. In addition to the samples taken to show the distribution of hydrocarbons in bay-bottom muds, samples of mud from experimental field stations and aquaria were extracted. Water samples, samples of oysters, and samples of plant remains were also studied.

Many mud samples were divided into two parts. One part was sent to the chemistry laboratory at the Agricultural and Mechanical College of Texas at College Station. These samples were analyzed for "non-volatile hydrocarbons" by Potts and assistants. The other aliquot, frozen and packed in dry ice, was sent to the Horvitz Laboratories at Houston, Texas, to be analyzed for volatile hydrocarbons in the range from methane to "pentane and heavier." This latter category was indefinite and overlapped the range of the non-volatile extractions. Although the extracted "hydrocarbons" were referred to as fractions of petroleum (methane, butane, etc.), it was not known whether they were actually petroleum hydrocarbons or whether they were fractions of oils from diatoms or other plankton organisms, or from higher plants.

Possible Sources of Pollution

If pollution were continuous and widespread enough to destroy oysters in a wholesale manner, it had to have a continuous source, and be on a large scale. Only the

"bleedwater," the brine effluent from salt-dome oil fields, was produced in quantity and on a continuous basis. For this reason, bleedwater measurements and analyses had priority in chemical studies.

In the Barataria Bay area, bleedwater was produced at several fields. The most important of these, in the 1940–47 period, were in the fresh and brackish-water areas north of the oyster-producing salt marsh zone (Delta Farms, Lake Salvador, LaFitte, Bayou Barataria, Lake Hermitage, Stella, and Bayou LeFleur). In the oyster-producing zone were Lake Washington, Queen Bess, and Bay DuChene fields. Queen Bess was a single-well affair, and the Bay DuChene field was just starting development. The latter is now a widespread field. To the east were the English Bay and Venice fields. The latter was in fresh water but comparatively close to oyster production. West of Bayou Lafourche were Lake Barre, Caillou Island, Bay Ste. Elaine, Lake Pelto, Dog Lake, and some other fields of minor importance. All of these were in oyster-producing bays. Straddling Bayou Lafourche was the Leeville field, and farther up, the Golden Meadow production. Locations of these fields are shown in Fig. 21.

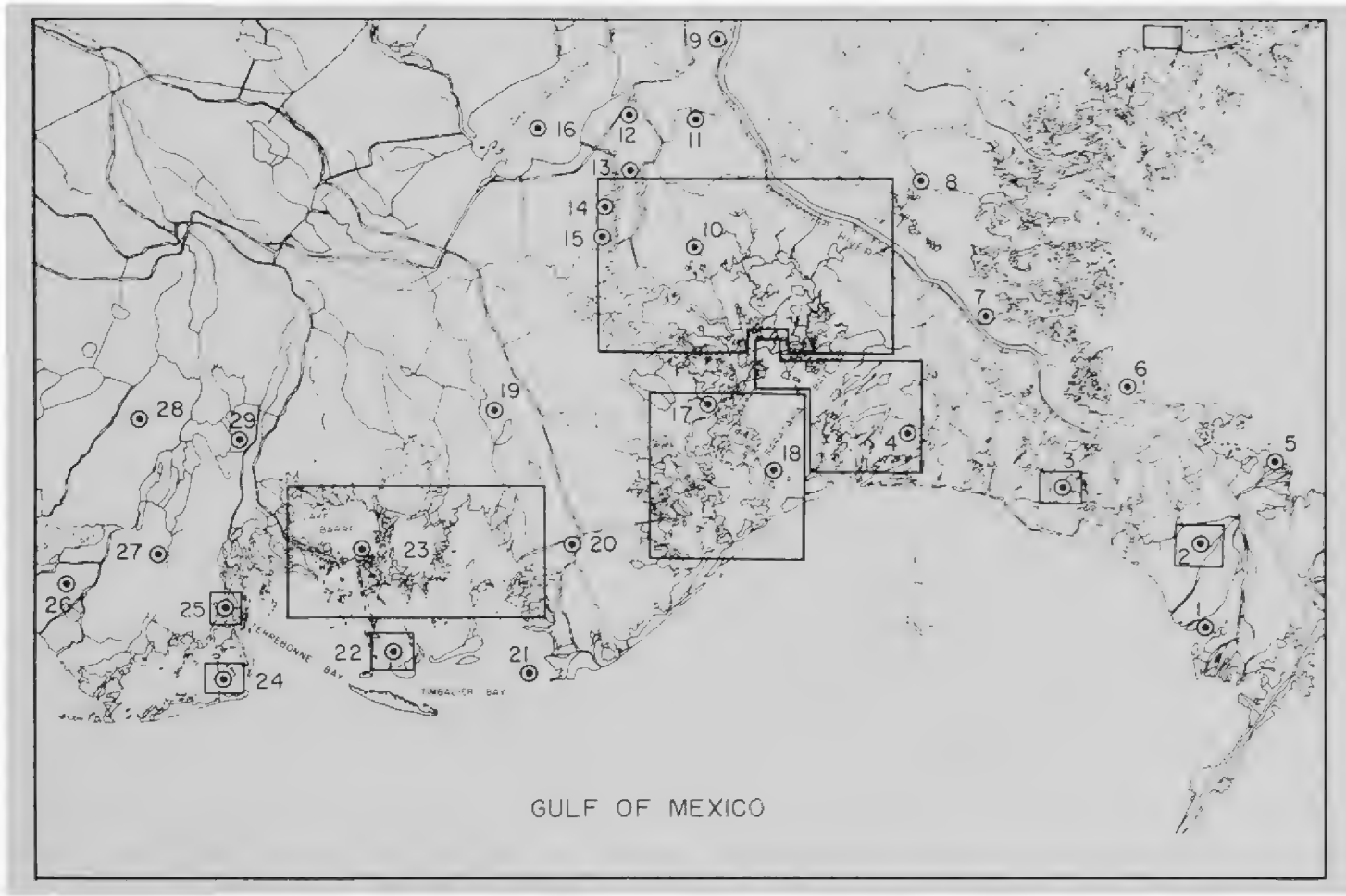

FIG. 21. Map showing locations of the principal oil fields in the area of Project 9 and Project 23 studies. Fields are as follows: 1. West Bay; 2. Venice; 3. English Bay (Bastian); 4. Lake Washington; 5. Grand Bay; 6. Quarantine Bay; 7. Potash; 8. Kenilworth; 9. Stella; 10. La Fitte; 11. Bayou La Fleur; 12. Barataria; 13. Bayou Perot; 14. Delta Farms; 15. Little Temple; 16. Lake Salvador; 17. Bay du Chene; 18. Queen Bess Island; 19. Golden Meadow; 20. Leeville; 21. Timbalier Bay; 22. Caillou Island; 23. Lake Barre; 24. Lake Pelto; 25. Bay Ste. Elaine; 26. Dog Lake; 27. DuLarge; 28. Dulac; 29. Lapeyrouse. Large enclosed areas containing oil fields define the areas of Figs. 22 to 31. The open rectangle in the upper right of the map encloses the area from which control cores were taken.

Some of the oil fields named were small and produced no bleedwater in the period 1941 to 1947. Some produced large amounts. In the Barataria basin, the LaFitte field produced about 7600 barrels daily in March, 1947 (Hewatt, 1950), and a mean of about

3000 barrels from 1941 to 1946 (Jensen, 1948a), and Lake Salvador produced about 2400 barrels daily in 1949. All others produced much less or none at all. The Venice field (Tidewater) in 1948 produced daily about 900 barrels of bleedwater which was evaporated in flare pits.

In the Terrebonne Bay area, some of the fields produced very considerable amounts of bleedwater. The largest production was by the Lake Barre field which produced an average of 6603 barrels per day in the period June 1, 1947, to June 1, 1949 (Menzel and Hopkins, 1951). Some fields produced relatively small amounts; for example, at Bay Ste. Elaine the amount averaged 258 barrels per day (Menzel and Hopkins, 1953). Other fields ranged between these figures. For a complete analysis see Jensen (1950).

Bleedwaters contain some oil, the amounts varying widely. Jensen, in the paper cited above, concluded that the bleedwaters discharged into the open bays (contrasted with flare pit discharge) contained on the average less than 30 ppm of petroleum hydrocarbons.

Accidental losses of crude petroleum occur during drilling, storage, and transportation operations. Amounts vary from a fraction of a barrel to thousands of barrels. The exact amounts are never known. Careful studies of the effects and duration of some of these losses have been made and will be presented in another section.

Natural Hydrocarbons in Muds

To establish the level at which extractable hydrocarbons occur naturally in muds, certain "control" samples were taken at various times and analyzed for "hydrocarbon" content. The various places from which these control samples were taken were all free from the possibility of contamination by petroleum from production centers, but some were exposed to slight contamination by bilge oil from small boat traffic. Some samples were not exposed even to oil from this source.

Table 10 presents the data derived from the control samples. One set of these was taken at Halfmoon Lake in the Louisiana Marsh, St. Bernard Parish. Study of the Halfmoon Lake area indicated that there was no possibility that oil production could have contributed to the "hydrocarbons" of muds there. The range of "hydrocarbon" values was from 0.009 percent to 0.062 percent at Halfmoon Lake. A number of samples were taken on the sea side of the Eastern Shore of Virginia. There has never been oil production or exploration in all of that region. There the maximum and minimum values ranged from 0.004 to 0.058 percent, certainly not far from the Halfmoon Lake values. The two samples from Allen Lake (0.025 and 0.068) were not, as far as can be determined, subject to oil contamination from any source, nor were the samples from Nebraska (0.003 to 0.034 percent).

The "hydrocarbon" content of these control samples must be considered in assessing the meaning of "hydrocarbon" values found in the high oyster mortality areas of Louisiana. They show that (1) "unsaponifiable hydrocarbons" probably exist in muds anywhere one may wish to take a sample, (2) the values obtained from extraction vary widely and erratically, and (3) the recording in mud samples of substances identifiable by routine chemical procedures as "hydrocarbons" does not mean that these necessarily result from pollution caused by oil production or exploration.

Origin of Hydrocarbons in Muds

It was noted early in the investigations that when cores were taken in areas in which

TABLE 10

Unsaponifiable residues of control samples of mud from various sources

Location of samples	Number of cores (samples)	Unsaponifiable residue: percent of composite of samples	Pentane and heavier percent
Halfmoon Lake, La., 8–27–48	9	0.027	0.0000055
Halfmoon Lake, La., 8–11–49	12	0.062	0.0000055
Halfmoon Lake, La., 4–1–49	2	0.009	0.0000043
Halfmoon Lake, La., 4–1–49	5	0.017	.....
Revell's Bay, Va., Smith & Walker oyster beds, 10–13–47	1	0.019	.....
Revell's Bay, Va., Smith & Walker oyster beds, 10–13–47	1	0.058	.....
Revell's Bay, Va., 3–22–48	9	0.005	.....
Revell's Bay, Va., 3–22–48 (avg. 3 depth samples)	1	0.004	.....
Burton's Bay, Va., 3–22–48	9	0.008	.....
Burton's Bay, Va., 3–22–48 (avg. 3 depth samples)	1	0.008	.....
G. Hopkins's wharf, North River, Va., 10–15–47	1	0.019	.....
Darling oyster beds, Hampton, Va., 10–17–47	5	0.013 (0.006–0.023)	.....
Mud Creek, Stratton, Nebr., 8–21–48	3	0.018 (0.007–0.034)	.....
Canyon, Stratton, Nebr., 8–21–48	2	0.004 (0.003–0.004)	.....
Frenchman River, Palisade, Nebr., 8–22–48	2	0.007 (0.006–0.007)	.....
Allen Lake, Bryan, Texas, 2–20–48	1	0.068	.....
Allen Lake, Bryan, Texas, 2–20–48	1	0.025	.....

the muds had a high percentage of organic matter, the "hydrocarbon" values appeared to be correspondingly high. The organic matter consisted of plant remains for the most part. Semi-peat deposits in which the stems and leaf fragments were clearly visible to the unaided eye are common throughout the Delta area. These deposits have their origin in the marsh vegetation. In the middle of the large bays, and in bottoms of deep bayous, these peaty layers as such may be absent. This probably is due to long exposure to oxidative processes or to scouring action of moving water where depths are comparatively great. Depths of twelve feet are uncommon in the Louisiana bays and bayous. Peaty layers are more common in the margins of small bays and bayous, and all marsh land is likely to be underlain by successive peaty strata, the top one of which is made up largely of living plant roots. In fresher waters of bayous which have growths of water hyacinth and alligator weed, the muds are often rich in plant remains.

Several studies were made to determine whether or not the apparent association of "unsaponifiable hydrocarbons" with plant remains was real. First, a bayou was selected which was characterized by heavy plant growth and deep muds filled with organic debris. This was Bayou Dupont, in its upper reaches and in two of its small tributaries. The sampled area was about 10 to 15 miles above the outlet of the bayou into northern Barataria Bay, isolated from any oil operations by drainage features, and in nearly fresh water. The locations of samples are shown in Fig. 22. The G and H series of samples were taken with an Ekman dredge. The J series was taken with a corer, two cores three feet deep being made into a composite and the third three-foot core being divided into top, middle, and bottom parts which were analyzed separately. The M series consisted

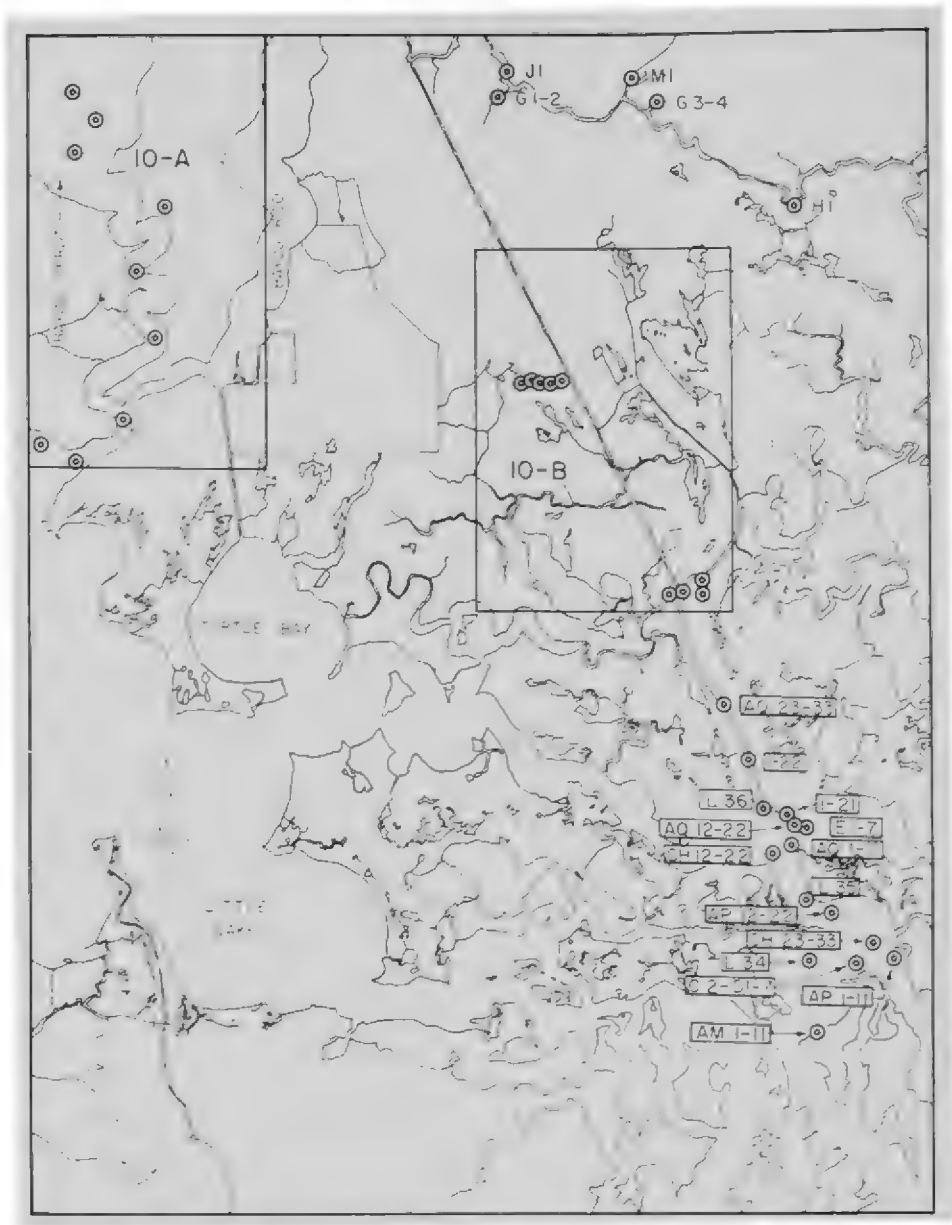

FIG. 22. Map showing locations of cores taken in the Bayou Perot (10-A) oil field, and LaFitte (10-B) oil fields and adjacent areas. Core locations in the northern part of the map are those taken on Bayou Dupont (Series G, H, J, M) to determine effect of plant debris.

of one three-foot core which was divided into six parts, each of which was analyzed separately. All data are presented in Table 11. These data show that the muds of these

TABLE 11

Cores taken in Bayou Dupont and in its small tributaries where heavy plant deposits occurred September 6 to October 20, 1947

Series	Location	Unsaponifiable residues, percent						
		Composite of cores	Depth cores					
			A*	B	C	D	E	F
G1†	Small bayou, tributary of Bayou Dupont	0.042	.....	.....	.....	.....	.....	.....
G2†	.....	0.006	.....	.....	.....	.....	.....	.....
J1‡	Bayou Dupont	0.040	0.005	0.032	0.021	.....	.....	.....
M1‡	Tributary of Bayou Dupont	.....	0.044	0.045	0.029	0.007	0.023	0.006
G3†	Tributary of Bayou Dupont	0.048	.....	.....	.....	.....	.....	.....
G4†	.....	0.077	.....	.....	.....	.....	.....	.....
H1†	Bayou Dupont	0.054	.....	.....	.....	.....	.....	.....

* A=top of core; other letters represent successively deeper layers down to about 40 inches.
† Samples taken by Ekman dredge.
‡ Samples taken by coring.

samples had a high "unsaponifiable hydrocarbon" content, and that the values varied widely. The indication was that in areas where the muds obviously had a high organic content, the "hydrocarbon" content also was high.

Further studies, which showed that the high values of "hydrocarbon" in muds were largely derived from plant remains, were made in the area of Lake Grande Ecaille. Here definite layers of peat alternate with silt layers and are easily separable on inspection of cores. A series of cores was taken at the eastern end of Lake Grande Ecaille and several more to the west of the lake. These were separated into peaty layers and silty layers.

Composite samples made up of peaty layers and of silty layers, respectively, were analyzed for "unsaponifiable hydrocarbons," separately for each series of cores. A total of 64 such cores was thus analyzed. The data are presented in Table 12. The locations of these samples are shown in Fig. 23.

TABLE 12

Comparison of unsaponifiable residues of carbon tetrachloride extracts in peaty layers and silty layers of cores taken in Lake Grande Ecaille, November 18, 1948, to December 3, 1949

Series	No. cores in series	Range, core length inches	Location of peat layers inches from top	Composite, unsap. residue of peat percent	Composite, unsap. residue of silt percent
Ba 1–5	5	36–40	middle 5–8	0.012	0.007
Ba 8–9	2	36–40	middle 12–18	0.013	0.006
Ba 12–14	3	32–36	top 4–12	0.074	0.008
Ba 17–19	3	37–40	top 4–6	0.018	0.003
Bq 1–6	6	32–40	middle 2–3	0.069	0.019
Bq 9–10	2	30–36	bottom 10 and middle 11	0.093	0.016
Bq 13–17	5	30–36	middle 2–6 and top 4	0.035	0.012
Bq 20–21	2	25–36	top 12 to top 20	0.053	0.019
Bq 24–27	4	20–25	top 2–8	0.005	0.014
Bu 1–17	17	20–40	top 10–22	0.046	0.002
Cn 1–6	6	.....	. . .	0.065	0.011
Bq 28–29	4	20–30	top 4–8	0.055	0.014
Totals	64	.....	Average	0.049	0.011

Study of these data shows that the "hydrocarbons" are associated with the plant remains, and it is believed that they are derived from the plants. The silty layers containing relatively small amounts of plant debris averaged 0.011 percent "unsaponifiable hydrocarbons" and the peaty layers averaged approximately four times as much (0.049 percent). In all sets of cores the peaty layers were significantly higher in "unsaponifiable hydrocarbons" than were the silty layers.

Two samples of dried water hyacinths from Allen Lake, Bryan, Texas, were analyzed for "unsaponifiable hydrocarbons" to determine the content of these substances in remains of plants which were recently alive. These plants had been dragged from the lake and piled on shore in an effort to rid the small lake of the pests. The two samples of water plants had respectively 0.096 percent and 0.214 percent "unsaponifiable hydrocarbons," considerably more than most mud samples on the average, and much more than any mud samples taken from the vicinity of oyster beds in open waters of bays.

Several core samples were taken in the Bay Ste. Elaine area in the vicinity of Texas Company wells drilled in exploration for sulphur. These cores were taken in the marshland, and the top four inches contained much living plant material in the form of roots.

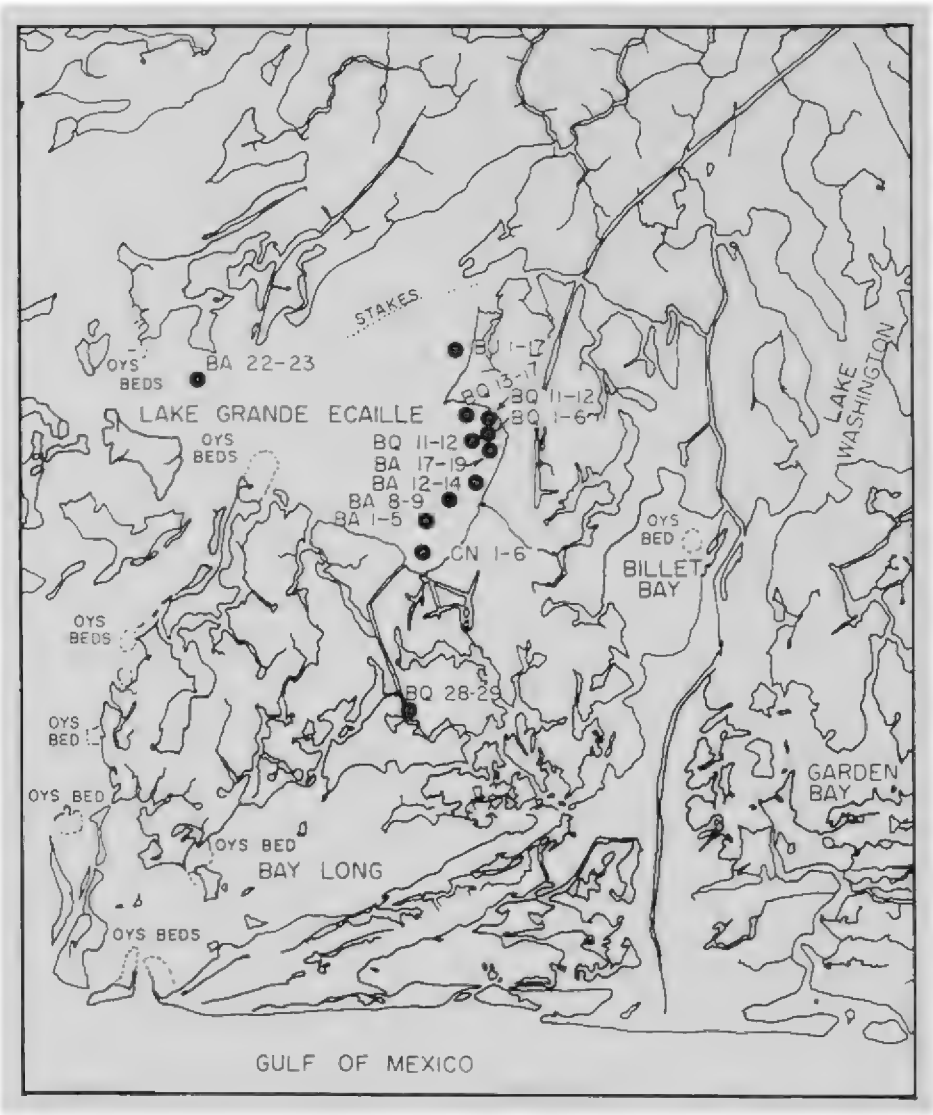

FIG. 23. Locations of cores tabulated in Table 12. These cores were used to compare "hydrocarbons" in peat with those in silts of the same cores. Most of these cores were taken in the east part of Lake Grande Ecaille.

When analyzed those parts of the cores containing the living plant material showed generally higher "unsaponifiable hydrocarbon" content than did the lower layers containing partially decomposed plant materials. The data are presented in Table 13.

TABLE 13

Cores taken in the marsh in the Bay Ste. Elaine area, January 20–21, 1948

Series	Percent unsaponifiable residue at depth		
	Top	Middle	Bottom
TM 1	0.157	0.020	0.041
UM 13	0.081	0.019	0.007
UM 2	0.058	0.014	0.016
TM 24	0.031	0.005	0.005
Mean	0.082	0.014	0.017

It is concluded from these studies that living plants contain relatively more "hydrocarbon" than do plant remains which have been partially oxidized.

The data presented here should be compared with the data in Table 25 showing "hydrocarbons" at different depths in cores from oyster-producing areas of Barataria Bay and adjacent waters. Most of the latter cores were taken around the margins of the bay (where the oyster leases are generally located), and in open waters for the most part. The Barataria Bay values are obviously lower than those associated with heavy

deposits of plant debris and living plant tissue, as in the Bay Ste. Elaine marsh cores. Table 24 shows that values of samples from Bay Ste. Elaine field also are generally lower than in these marsh cores.

Hydrocarbons in Muds and Their Relation to Oyster Mortality

It was reasoned that, if the hydrocarbons in muds were related to oyster mortality, there should be a significant correlation of high mortality and high "hydrocarbon" content. To determine whether such a relation existed, cores were taken (1) in an area where mortality had been found to be low and (2) where mortality had been observed to be high. The Bay Chene Fleur area was chosen as representative of an area where the mortality was low. Two fourteen-month checks showed the oyster mortality to be about 30 percent at this station (Mackin and Wray, 1949, 1950). At Sugar House Bend and Bayou Rigaud in lower Barataria Bay the losses had been similarly established at a level of approximately 80 percent in the same periods. Cores were taken in the general vicinity of each of these stations, and analyzed for unsaponifiable hydrocarbons. The data are presented in Table 14.

Table 14

Unsaponifiable residues in muds from low and high mortality areas

Series	Unsaponifiable residue, percent				Pentane-heavier percent
	Composite of cores	Depth core			
		Top	Middle	Bottom	
Chene Fleur Station Area (annual mortality rate about 30 percent), October 18, 1947 to March 24, 1949					
Ak 1–11	0.006	0.008	0.004	0.006	0.0000212
K 1–11	0.009	0.016	.	.	.....
Bf 12–22	0.005	0.005	0.006	0.006	0.0000007
Bg 12–22	0.005	0.016	0.024	0.012	0.0000021
Averages	0.006	0.011	0.011	0.008	0.0000080
Sugar House Bend and Bayou Rigaud (annual mortality rate about 80 percent), June 2, 1948, and November 18–19, 1949					
Cl 1–11	0.017	0.018	0.011	0.007	0.0000019
Ag 1–11	0.009	0.015	0.007	0.004	0.0000051
Ag 12–22	0.015	0.010	0.009	0.006	0.0000068
Ck 12–22	0.003	0.003	0.004	0.002	0.0000009
Averages	0.011	0.011	0.008	0.005	0.0000037

With one exception all values in both high and low mortality areas were low. One composite sample in the low mortality area had a "pentane and heavier" fraction of about 0.2 ppm which could indicate the presence of petroleum hydrocarbons in small amount, although the "unsaponifiable hydrocarbon" content was not above the average. On the whole, the data indicate that there was no significant difference in "hydrocarbon" content of the muds in the two areas.

Petroleum Hydrocarbons Around Bleedwater Outlets

The studies presented above show that "hydrocarbons" found in muds of most areas where oysters grow are of plant origin. However, hydrocarbons of undoubted petroleum origin were found in muds in the vicinity of some oil field bleedwater outlets.

Careful studies were made in and near oil fields in the marsh to the north, in fresh-water areas above the oyster-producing areas where mortality from pollution was claimed. Wherever indications of oil were found, the movement of hydrocarbons away from the effluent outlets was traced, if possible. These studies showed that wherever large amounts of bleedwater were discharged into confining canals and bayous, there were considerable accumulations of oil in the bottom muds. Depending on the degree of confinement of the bleedwater and the amount of the discharge, various degrees of pollution occurred at the site of the discharge. Away from the point of discharge very rapid disappearance of the oil took place. The heaviest concentration of oil at a bleedwater outlet was found in a canal at the LaFitte field. This area was studied closely. The LaFitte field is not in oyster-producing territory but is situated seven to eight miles to the northwest of Barataria Bay and about five miles from the nearest oyster-producing area. The point of bleedwater discharge was in an access canal, which in turn connects with Dupree Cut, a dredged channel running southeast through the marshes for about two and one-half miles. At its southeast end, Dupree Cut joins Bayou Cutler. Bayou Cutler then continues roughly to the southeast for another two and a half to three miles, to its juncture with Bayou St. Denis. Oysters are grown in Bayou St. Denis. This bayou continues for another three miles to its junction with Barataria Bay. The relations are shown in Fig. 24.

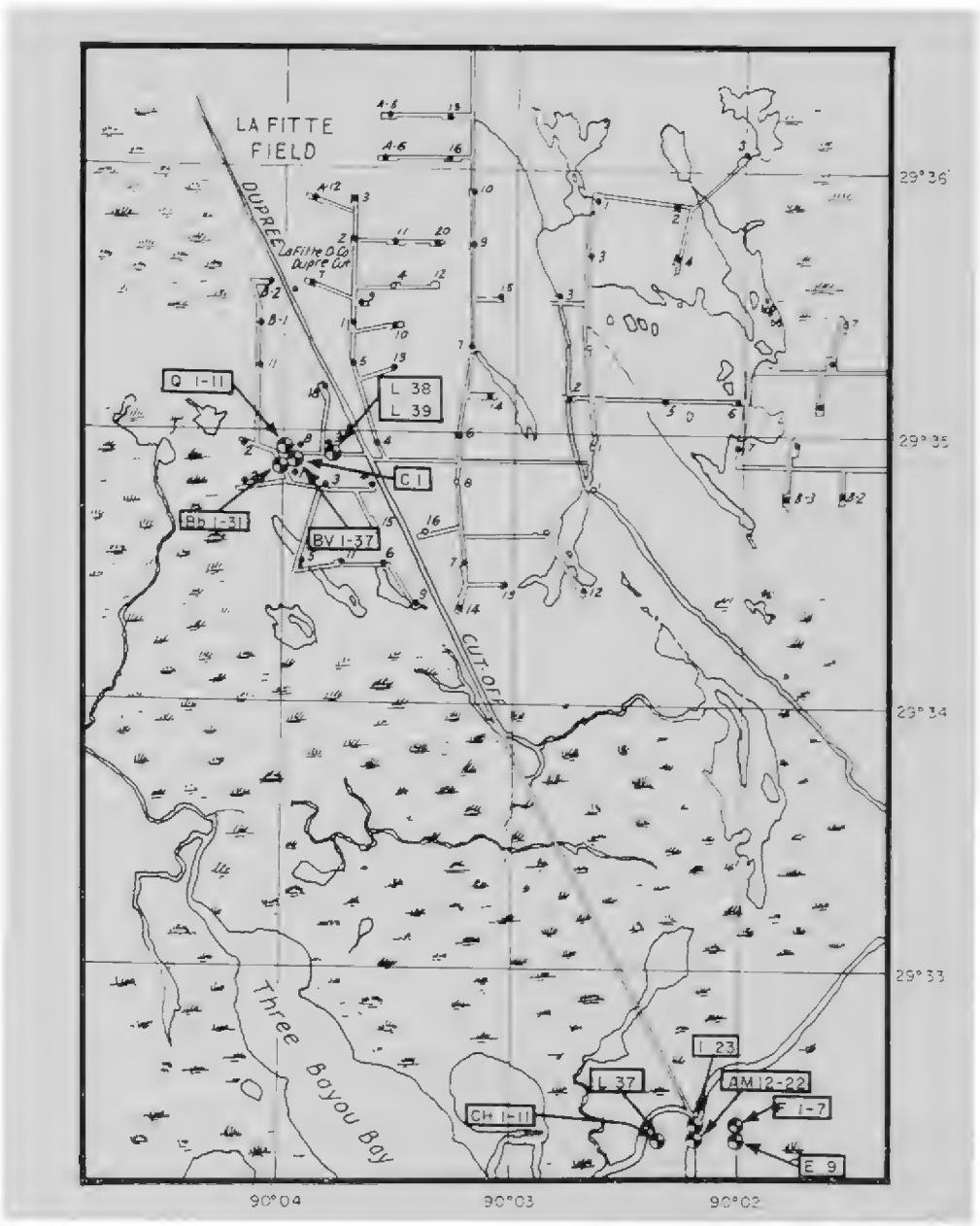

FIG. 24. Locations of cores used for hydrocarbon sampling in the LaFitte oil field and at the south end of Dupree cut-off canal. For others of this series see Fig. 22.

LaFitte is a large oil field and produced a large amount of bleedwater from the separators, all going into the access canal at Camp LaFitte. The average daily production was around 3000 barrels per day in the period 1941 to 1946 inclusive (Jensen 1948a). All of this entered the access canal at one point, referred to here as the "bleedwater outlet," or "BW outlet." It was assumed that net movement of the water was generally seaward and that dilution occurred as water from the access canal mixed with that in Dupree Cut and successively with each water body which added to the stream. Currents in Dupree Cut are feeble and dictated largely by wind (Lynch, 1948). Lynch calculated that dilution of bleedwater with water in Dupree Cut reduced the concentration to less than 0.7 percent at the juction of the access canal with the cut.

TABLE 15

"Hydrocarbon" content of cores taken at LaFitte Field and below in Bayou Cutler to Barataria Bay, August 13, 1947, to November 15, 1949

Series	Position, distance from bleedwater source	Unsaponifiable residue, percent					Pentane and heavier percent
		Composite of cores	Range of values	Depth in core			
				Top	Middle	Bottom	
Bb 1–31	At BW outlet in canal at camp	2.526	2.244 to 2.819	.....	.....	.....	0.0010300
Bv 1–37	At BW outlet in canal at camp	5.80	.....	.....	.....	.....	0.0014500
Q 1–10	At BW outlet in canal at Camp LaFitte	0.570	0.011 to 4.800	20.320	1.640	0.060	0.0004740
L 38	75 ft below BW outlet in canal at LaFitte	0.041	.....	.....	.....	.....	0.0005930
L 39	In canal at LaFitte camp warehouse, about 1400 ft below BW outlet	0.044	.....	.....	.....	.....	0.0000173
C 1	Same	0.101	.....	.....	.....	.....	0.0001040
Am 12–22	2½ miles; junction of Dupree Cut and Bayou Cutler	0.007	0.006 to 0.016	0.045	0.008	0.005	0.0000044
Fi 1–7	Just west of junction Dupree Cut and Bayou Cutler	0.033	0.024 to 0.048	.....	.....	.....	.....
E 9	Junction Dupree Cut and Bayou Cutler (west bank of bayou)	0.034	.....	.....	.....	.....	.....
L 37	Near west bank of Bayou Cutler; 100 yards from junction Dupree Cut	0.012	.....	.....	.....	.....	0.0000040
Ch 1–11	200 yards down Bayou Cutler from junction with Dupree Cut	0.023	.....	0.014	0.012	0.024	0.0000048
I 23	50 ft from east bank Bayou Cutler at junction with Dupree Cut	.....	.....	0.011	0.017	0.009	.....
Aq 23–33	About 4¼ miles S of BW outlet in Bayou Cutler	0.008	0.004 to 0.014	0.020	0.003	0.003	0.0000040
I 22	About 4⅔ miles S of BW outlet in Bayou Cutler, 45 ft from west bank	.....	.....	0.008	0.029	0.010	.....
L 36	5½ miles from BW outlet; in Bayou Cutler ¼ mile above Bayou St. Denis	0.017	.....	.....	.....	.....	0.0000026

TABLE 15—Continued

"Hydrocarbon" content of cores taken at LaFitte Field and below in Bayou Cutler to Barataria Bay, August 13, 1947, to November 15, 1949

Series	Position, distance from bleedwater source	Unsaponifiable residue, percent: Composite of cores	Unsaponifiable residue, percent: Range of values	Unsaponifiable residue, percent: Depth in core: Top	Middle	Bottom	Pentane and heavier percent
Aq 12–22	5¾ miles from BW outlet above junction Bayou Cutler and B. St. Denis in B. Cutler	0.009	0.005 to 0.014	0.031	0.009	0.004	0.0000060
I 21	5¾ miles from BW outlet at junction B. Cutler and B. St. Denis; 45 ft from east shore	.....	.....	0.010	0.003	0.010	.....
E 1–6	5¾ miles from BW outlet at junction of B. Cutler and B. St. Denis opposite light	0.023*	0.016 to 0.032	.....	.....	.....	.....
Aq 1–11	Bayou St. Denis ¼ mile below junction with Bayou Cutler; 6 miles from BW	0.007	0.002 to 0.014	0.040	0.006	0.002	0.0000060
Ch 12–22	Bayou St. Denis just below junction B. Cutler; 6 miles from BW outlet	0.007	.....	0.017	0.009	0.006	0.0000012
L 35	7 + miles below BW outlet B. St. Denis east bank at cut into Mud Lake	0.016	.....	.....	.....	.....	0.0000006
Ap 12–22	7 + miles below BW outlet B. St. Denis	0.013	0.006 to 0.016	0.025	0.014	0.011	0.0000117
Ch 23–33	8 miles from BW outlet B. St. Denis	0.028	.....	0.044	0.013	0.011	0.0000058
L 34	8 + miles from BW outlet; west side mouth of Mud Lake 75 ft from shore	0.013	.....	.....	.....	.....	0.0000030
C 2	8 + miles from BW B. St. Denis south Mud Lake entrance	0.028	.....	.....	.....	.....	0.0000033
D 1–7	8 + miles from BW outlet at south Mud Lake entrance	0.010	0.008 to 0.036	.....	.....	.....	.....
Ap 1–11	8 + miles below BW B. St. Denis near east side	0.006	0.003 to 0.008	0.012	0.004	0.007	0.0000055
Am 1–11	South shore of Mud Lake 8½ miles from BW outlet	0.013	0.007 to 0.019	0.086	0.020	0.009	0.0000024
W 1–11	Mouth of B. St. Denis at North Pt. 9 miles below outlet	0.013	0.007 to 0.016	0.010	0.006	0.023	0.0000022
Ci 1–11	Mouth of Bayou St. Denis 9 miles below BW outlet	0.009	.....	0.005	0.005	0.010	0.0000013

* Not composite, but mean of values for 6 cores.

Table 15 shows the "hydrocarbon" content of muds in the access canal at the bleedwater outlet and at various points seaward to the junction of Bayou St. Denis with Barataria Bay. The first three sets of values given were from samples taken at the outlet. These show large amounts of extractives in the mud bottom in the canal at the point of

discharge. Attention is called especially to the 10 cores of set Q 1–10. The range of values in this circular pattern is remarkable (0.011 to 4.800 percent). The depth core, that is, the one in which samples were analyzed at top, middle, and bottom, showed an even higher value in the surface four inches (20.320 percent). Attention is called to the "pentane and heavier" fractions reported for these three sets (Bb, Bv, and Q) at the bleedwater outlet. This fraction averaged about 10 ppm. It has been noted that the "pentane and heavier" fraction is more reliable than others in drawing a picture of relative amounts of petroleum hydrocarbons in the muds. Values of less than 0.1 ppm are found widely distributed in muds of Louisiana (Jensen and Potts 1953). Any values sensibly above 0.1 ppm may be taken to mean that petroleum oil is probably present, and if much higher than that figure it is nearly certain that such is the case. The "pentane and heavier" fractions at the bleedwater outlet ranged, approximately, from 5 to 15 ppm, probably indicating high levels of petroleum accumulation in the muds.

The oil at the LaFitte bleedwater outlet showed very rapid decrease with distance away from the outlet. At 75 feet above 40 percent had disappeared, and at 1400 feet 94 percent reduction had occurred. These stations were in the access canal. At the lower end of Dupree Cut, the "hydrocarbon" values indicate that no measurable quantity of oil was present in the muds, and from there to Barataria Bay there is no indication of pollution as shown by the table.

As a means of illustrating the disappearance of the oil with distance from the bleedwater outlet a table was compiled, based on the values of the composite samples analyzed for "pentane and heavier" fractions. The value obtained by averaging the values of the three samples at the bleedwater outlet was rated at 100. The values of the sample at 75 feet, the two at 1400 feet, and the one at the junction of Dupree Cut with Bayou Cutler (2½ miles distant, approximately) were calculated in percentage of the values at the bleedwater outlet. Table 16 shows the results.

It is not believed that the rapid reduction in amount with distance, shown in the table, resulted from dilution alone. In a canal like the access canal where the water movement is so weak as to be almost negligible, dispersal by currents must be slow, a conclusion borne out by the failure (see section on hydrocarbons in water) to find significant amounts of oil in the water or to find surface slicks at any significant distance from the outlet. Slicks are inevitable where oil in quantity is carried by water, because of the low specific gravity of the oil. Yet a rough calculation from the values found in samples shows that not much more than 10 or 12 barrels of oil were in the muds around the bleedwater outlet at any one time. But the bleedwater has discharged at this point for years, and it was apparent to the investigators that the total or cumulative amount of oil was very much larger than that present at the time of sampling. In other words, it

TABLE 16

Disappearance of oil in bottom muds with distance from bleedwater outlet at the LaFitte Field: "Pentane and heavier" fractions

Distance of samples from BW outlet	Percent of hydrocarbons present based on a value of 100 at the outlet
At BW outlet	100.00
75 ft from BW outlet	60.22
1400 ft from BW outlet	6.16
2½ miles from BW outlet	0.45

did not all accumulate; some oil disappeared in the course of time. The disappearance of oil is discussed in detail below.

At other fields, concentrations of petroleum in muds at the bleedwater outlets were less than those found at LaFitte. At the Delta Farms field there was oil in the muds of Bayou Perot and the "pentane and heavier" fractions showed that there was a low-level persistent pollution extending to the lower limits of the southerly parts of the several small oil fields, elements of which extend for 2½ miles along Bayou Perot. Below this area, "hydrocarbons" in amounts attributable to petroleum contamination were not found. The data are presented in Table 17. No part of the Delta Farms field is in or near oyster-producing territory and Bayou Perot is nearly fresh water. The locations of the samples listed in Table 17 are presented in Fig. 25.

At the Lake Hermitage field in the fresh-water marshes to the northeast of Barataria Bay, no bleedwater was produced in the period 1941 to 1947, and there were only two producing wells in 1947. The amounts of "hydrocarbons" found in muds in this field are presented in Table 18. These values should be compared with those of oyster-producing areas of Barataria Bay (Table 25). The locations of these samples are shown in Fig. 26.

At the English Bay field, which is in oyster-producing territory, only one set of cores among the many taken indicated that there were probably petroleum hydrocarbons in the muds. This set (Ce 1–11) was taken in a company canal roughly 400 to 500 feet from the bleedwater burning pit. Other cores, including those taken much closer, showed "unsaponifiable hydrocarbon" and "pentane and heavier" fractions so low that they must be considered to be negative for petroleum. Cores were taken at varying distances from the bleedwater burning pit into and across English Bay to the east, but no indi-

TABLE 17

"Hydrocarbons" in muds in Bayou Perot in and below the Delta Farms Field
August 13, 1947, and August 17 and 31, 1948

Series	Approximate distance from nearest well or BW outlet	Unsaponifiable residue, percent					Pentane and heavier percent
		Composite of cores	Range of values	Depth in core			
				Top	Middle	Bottom	
At 34–44	Zero (in canal from main field) Humble Co.	0.066	0.048 to 0.289	0.109	.....	.....	0.0000043
At 23–33	Zero at bleedwater outlet T.T. Co. Delta Farms Field	0.046	0.021 to 0.825	0.079	0.106	0.072	0.005780
At 12–22	About ⅕ mile below nearest well	0.080*	0.003 to 0.300	0.026	0.069	0.077	0.0000096
At 1–11	1 mile below field	0.033	0.023 to 0.078	0.023	0.042	0.053	0.0000108
Ar 34–44	2 miles below field	0.045	0.041 to 0.059	.....	.....	0.059	0.0000123
Ar 23–33	2½ miles below field	0.086	0.002 to 0.025	0.002	0.012	0.005	0.0000039
Ar 12–22	3½ miles below field	0.027	0.009 to 0.050	0.050	0.018	0.009	0.0000052
Ar 1–11	4 miles below field	0.002	0.006 to 0.015	0.015	0.005	0.009	0.0000029
C 3	5 miles below field	0.036	.....	.....	.....	.....	0.0000032

* Not a composite, for which data were lost, but mean of all cores except the one reserved for depth analysis.

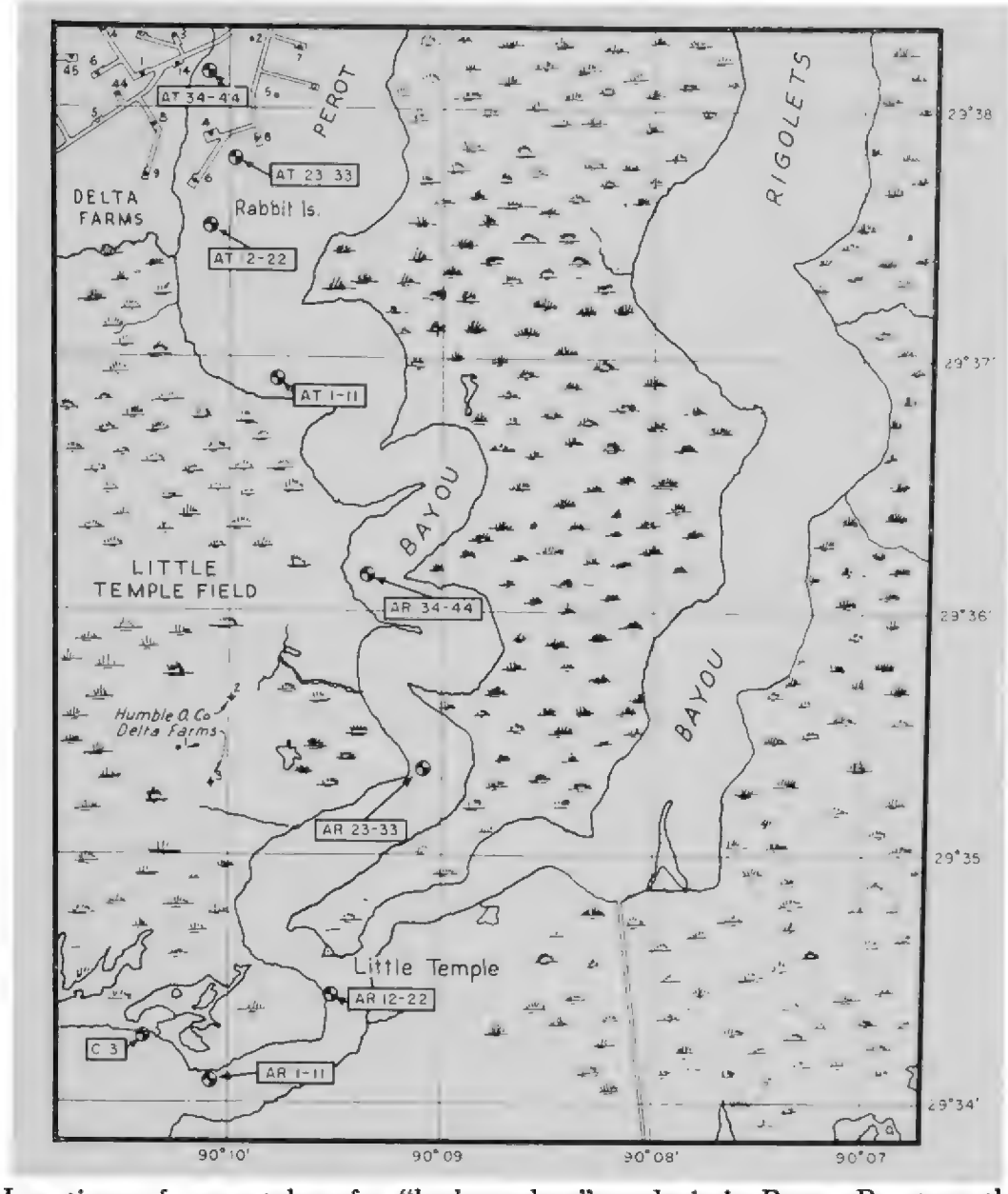

FIG. 25. Locations of cores taken for "hydrocarbon" analysis in Bayou Perot, at the Delta Farms and Little Temple oil fields.

cation of pollution was found in these. Table 19 reports the "hydrocarbon" values found in making this study. The locations where core samples were taken for "hydrocarbon" measurement in the English Bay field are shown in Fig. 26a.

The conditions at the Lake Barre Field of the Texas Company above Terrebonne Bay deserve special comment because this was the site of extensive studies on the effect of bleedwater on oysters which are reported in another section. Here oysters grow well even under the well rigs and on the pilings of the cat walks and the bulkheads. Employees of the Texas Company regularly pry these oysters loose and use them for food. They are generally very good oysters when fat in winter. Some oysters grow within a few feet of the bleedwater outlets, but they are noticeably less in number here.

As stated above, bleedwater production at the Lake Barre field averaged 6603 barrels per day, with a maximum production of 9120 barrels per day. This bleedwater is discharged through two outlets set within a few feet of each other. The discharge is into relatively open water. There is no land within miles toward the north, and only little land in any direction. The only nearby "land" is in the form of broken-up marsh islands.

The data show that the mud samples taken at the bleedwater outlet at Lake Barre almost certainly contained some petroleum hydrocarbons (Table 20). However, the "pentane and heavier" fractions showed only 1.17 ppm in the circular pattern of cores at

TABLE 18

Unsaponifiable residues of muds in and around the Lake Hermitage Field. All distances are approximations in nautical miles. October 18, 1947, to August 4, 1949

Series	Location and distance from separators (Note: no BW produced here; only distillate)	Unsaponifiable residue, percent				Pentane and heavier percent
		Composite of cores	Depth core			
			Top	Middle	Bottom	
Bc 12–22	Junction of Wilkinson and Hermitage Bayous; in field	0.053	0.070	0.113	0.026	0.0000034
Bc 1–11	Bayou Wilkinson at end of canal from separator 1250 ft from BC 12	0.014	0.040	0.009	0.009	0.0000020
Bc 23–33	Bayou Wilkinson at junction Racquette Bayou 1.7 miles below separator	0.016	0.038	0.017	0.024	0.0000021
Bd 1–11	Wilkinson Bayou 1.8 miles below separator	0.021	0.025	0.008	0.015	0.0000019
Bd 12–22	Upper Petit Bay Chene Fleur; 2 miles from separator	0.008	0.029	0.017	0.010	0.0000023
Bd 23–33	Wilkinson Bayou; about 2.7 miles below separator	0.016	0.038	0.017	0.024	0.0000021
Be 1–11	Wilkinson Bayou; 3 miles below separator	0.016	0.008	0.015	0.010	0.0000016
Be 23–33	Wilkinson Bayou; 3¼ miles below separator	0.012	0.038	0.020	0.016	0.0000025
Be 12–22	Wilkinson Bayou; ¾ mile below separator	0.008	0.005	0.011	0.013	0.0000006
Bt 1–11	Wilkinson Bayou; 3.6 miles below separator	0.013	0.009	0.015	0.009	0.0000011
Bt 23–33	Upper Bay Chene Fleur at junction with N. Bayou; 3.8 miles below separator	0.005	0.005	0.006	0.006	0.0000007
Bt 12–22	Bay Wilkinson 4.5 miles below separator	.....	0.022	0.035	0.029	0.0000032
Ak 1–11	Southern part of Wilkinson Bay; 5.8 miles below separator	0.006	0.008	0.004	0.006	0.0000212
K 1–11	Wilkinson Bayou; 6 miles below separator	0.009	0.016	.....	.....	.....
Bg 12–22	Wilkinson Bayou; 6.5 miles below separator	0.012	0.008	0.009	0.006	0.0000032

the bleedwater discharge, and at 50 feet south only 0.12 ppm. At greater distances there was no indication that petroleum hydrocarbons were present in the muds. The locations of these samples are shown in Fig. 27.

The great difference in the concentration of petroleum hydrocarbons at Lake Barre and at LaFitte is due to the fact that at Lake Barre the effluent passes into relatively open water with positive tidal current movement and a great expanse of moderately saline water for dilution.

The effect of burning pits is shown by studies at the Venice field of the Tidewater Oil Company. This field is situated far down the Mississippi Delta in nearly fresh water. It is also in the marsh and in a system of confining canals. A number of burning pits evaporate or burn hydrocarbons in the bleedwater from the separators. Excess salt water from the pits drains into the canals. An extensive series of mud samples strategically distributed around the entire field failed to show contamination with petroleum hydrocarbons from bleedwaters (Table 21). Fig. 28 shows the locations of samples listed in Table 21 which were taken in and around the Venice oil field.

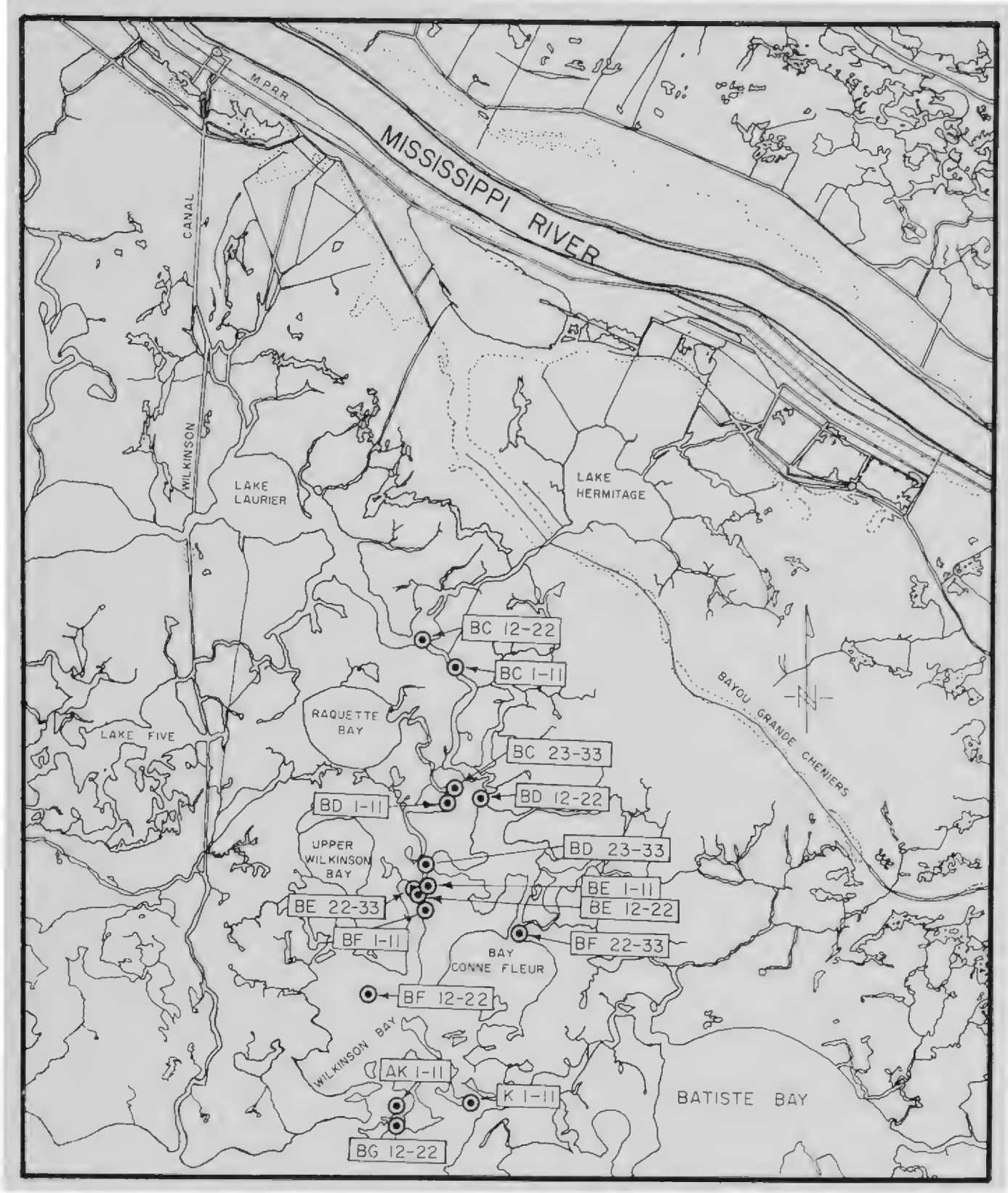

FIG. 26. Locations of cores taken for "hydrocarbon" analysis in Lake Hermitage field and southward.

At Caillou Island and Lake Pelto fields in lower Terrebonne Bay a different situation exists. Both fields are located in wide open water of high salinity. Bleedwater is discharged into the water directly. Cores taken at these bleedwater outlets failed to show any contamination of the muds within a few feet of the points where effluent was discharged. At Lake Pelto the bleedwater discharge averaged 1385 barrels per day from June 1, 1947, to June 1, 1949. At Caillou Island in the same period the average bleedwater production was about 5072 barrels daily. A large pass to the open Gulf is between and near these two fields, insuring a short flushing period. The values of the unsaponifiable residues for the Lake Pelto field are shown in Table 22, and those for Caillou Island in Table 23. Fig. 29 shows the locations of samples taken in the Lake Pelto field and Fig. 30 shows the locations of the samples in the Caillou Island oil field.

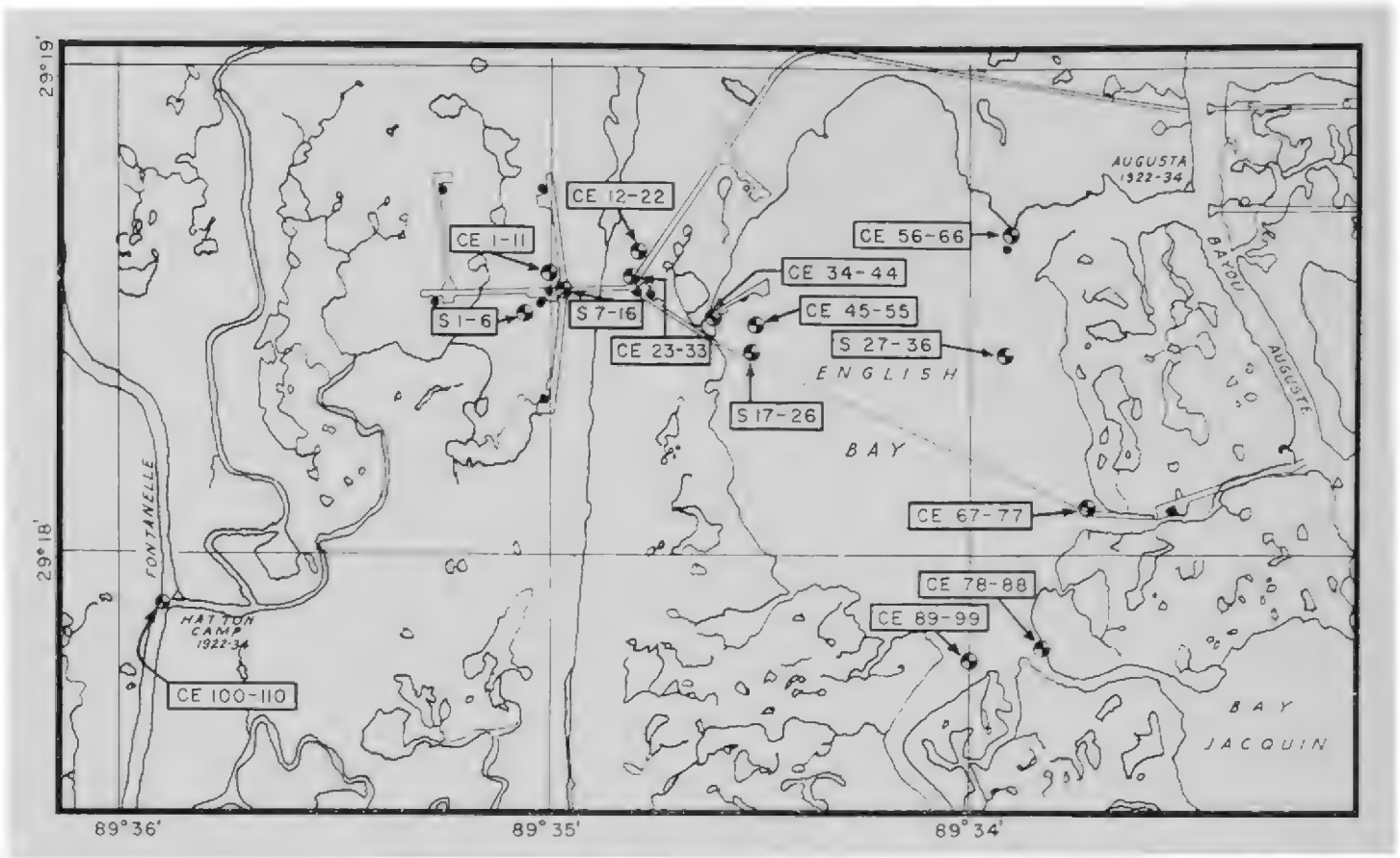

FIG. 26a. Locations of cores taken for "hydrocarbon" analysis in the English Bay field and adjacent areas.

TABLE 19

Unsaponifiable residues of muds at varying distances from the bleedwater burning pit in the English Bay Field, January 3, 1948 and October 21, 1949

Location; distance from BW outlet	Unsaponifiable residue, percent				
	Composite of cores	Depth in core			Pentane and heavier percent
		Top	Middle	Bottom	
In canal 100 ft from BW burning pit	0.037	....	.....	. .	....
At mouth of canal from BW pit, 300 ft	0.032	0.029	0.025	0.013	0.0000040
Just N of intersection of canals, 4–500 ft N of BW pit	0.048	0.258	0.023	0.057	0.0000615
In canal at "Y" ⅕ mile east of BW pit	0.036	0.046	0.054	0.022	0.0000021
About ¼ mile from BW pit in canal to NE	0.047	0.052	0.080	0.033	0.0000071
About ⅓ mile E and S of BW pit; west margin of English Bay	0.011	0.009	0.010	0.006	0.0000017
About ½ mile E of BW outlet in west side of English Bay	0.010	0.015	0.008	0.011	0.0000022
½ mile from BW outlet at mouth of canal at west margin of English Bay	0.013	0.025	0.021	0.008	.....
About 1 mile from BW outlet; east margin of English Bay	0.009	0.011	0.008	0.021	0.0000023
About 1 mile from BW outlet; northeast English Bay, in outlet to Bayou August	0.015	0.014	0.010	0.012	0.0000044
About 1¼ miles from BW pit; southwest English Bay	0.015	0.009	0.012	0.017	0.0000016
About 1½ miles from BW outlet; south part of English Bay	0.012	0.014	0.014	0.011	0.0000048
About 1½ miles from BW outlet; south part of English Bay	0.013	0.018	0.016	0.013	0.0000015
1½ miles SW of BW outlet in Bayou Fontanelle	0.018	0.021	0.019	0.015	0.0000027

TABLE 20

Unsaponifiable residues of muds at and around the bleedwater outlets at the Lake Barre Oil Field, December 22, 1947, September 1, 1948, and March 29–31, 1950

Series	Location	Unsaponifiable residue, percent					
		Composite of cores	Range of values	Depth in core			Pentane and heavier percent
				Top	Middle	Bottom	
(Hopkins)	At BW outlets	0.183	.....	.....	.....	.....	.....
P 13–22	At BW outlets	0.048	0.020 to 0.159	.....	.....	.....	0.0001170
Au 7–17	40 to 50 ft S of BW outlet	0.023	0.002 to 0.051	0.003	0.008	0.002	0.0000120
Cw 22	In channel ½ mile S	.....	.....	.....	.....	.....	0.0000011
Cu 77	In channel 1¼ miles S of BW outlet	.....	.....	.....	.....	.....	0.0000051
P 23–33	2¾ miles NW at entry to Lake Felicity	0.022	0.008 to 0.032	0.057	0.016	0.012	0.0000056

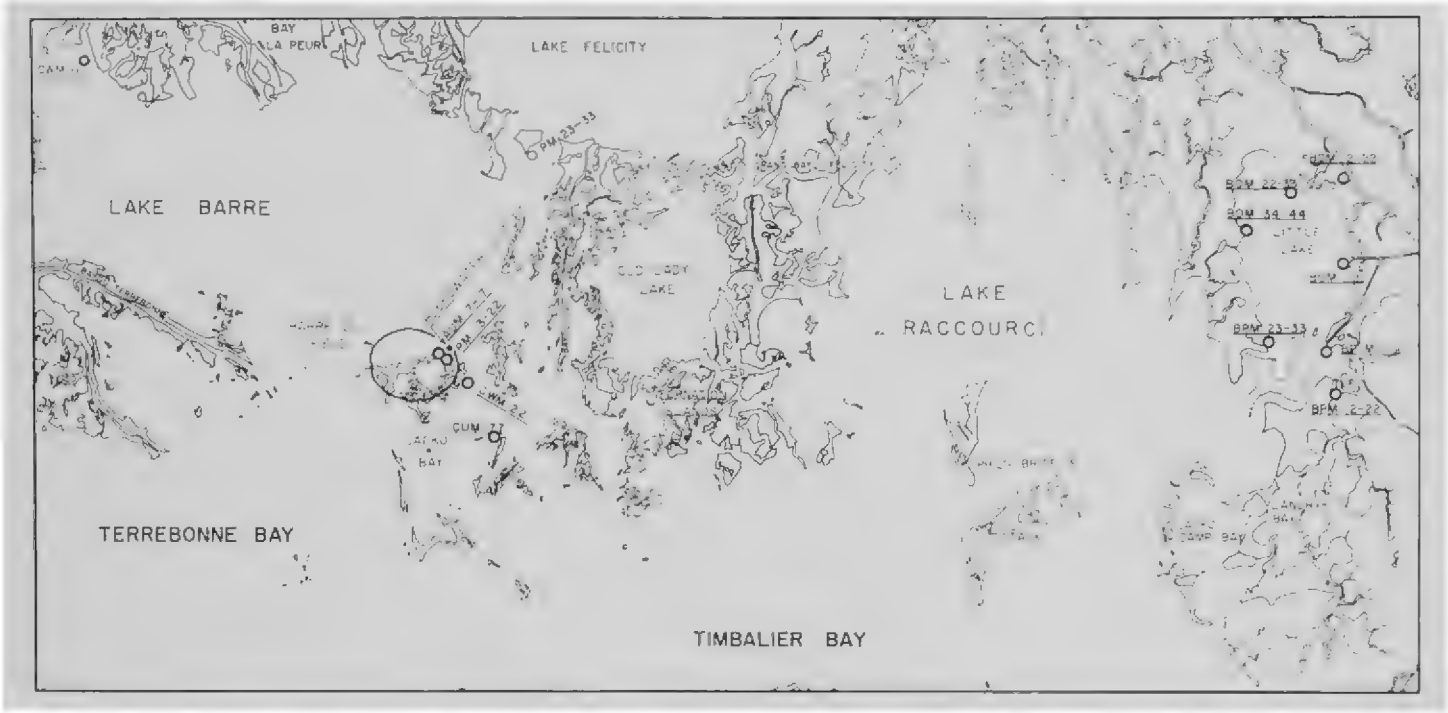

FIG. 27. Locations of cores taken for "hydrocarbon" analysis (1) in the area of the Lake Barre Oil Field, (2) in Little Lake.

TABLE 21

Unsaponifiable residues of extractives of muds near burning pits and in the canals of the Venice Oil Field, February 21–23, 1950

Series	Unsaponifiable residue, percent				
	Composite of cores	Depth in core			Pentane and heavier percent
		Top	Middle	Bottom	
Cr 34–43	0.018	0.010	0.011	0.022	0.0000017
Cr 23–33	0.018	0.032	0.007	0.007	0.0000045
Cr 12–21	0.032	0.073	0.021	0.013	0.0000026
Cr 1–10	0.017	0.057	0.025	0.017	0.0000032
Cs 12–21	0.009	0.006	0.009	0.007	0.0000032
Cs 1–11	0.008	0.007	0.005	0.002	0.0000028
Ct 12–21	0.008	0.009	0.012	0.006	0.0000022
Ct 23–32	0.008	0.011	0.012	0.015	0.0000057
Ct 34–43	0.011	0.014	0.007	0.007	0.0000031

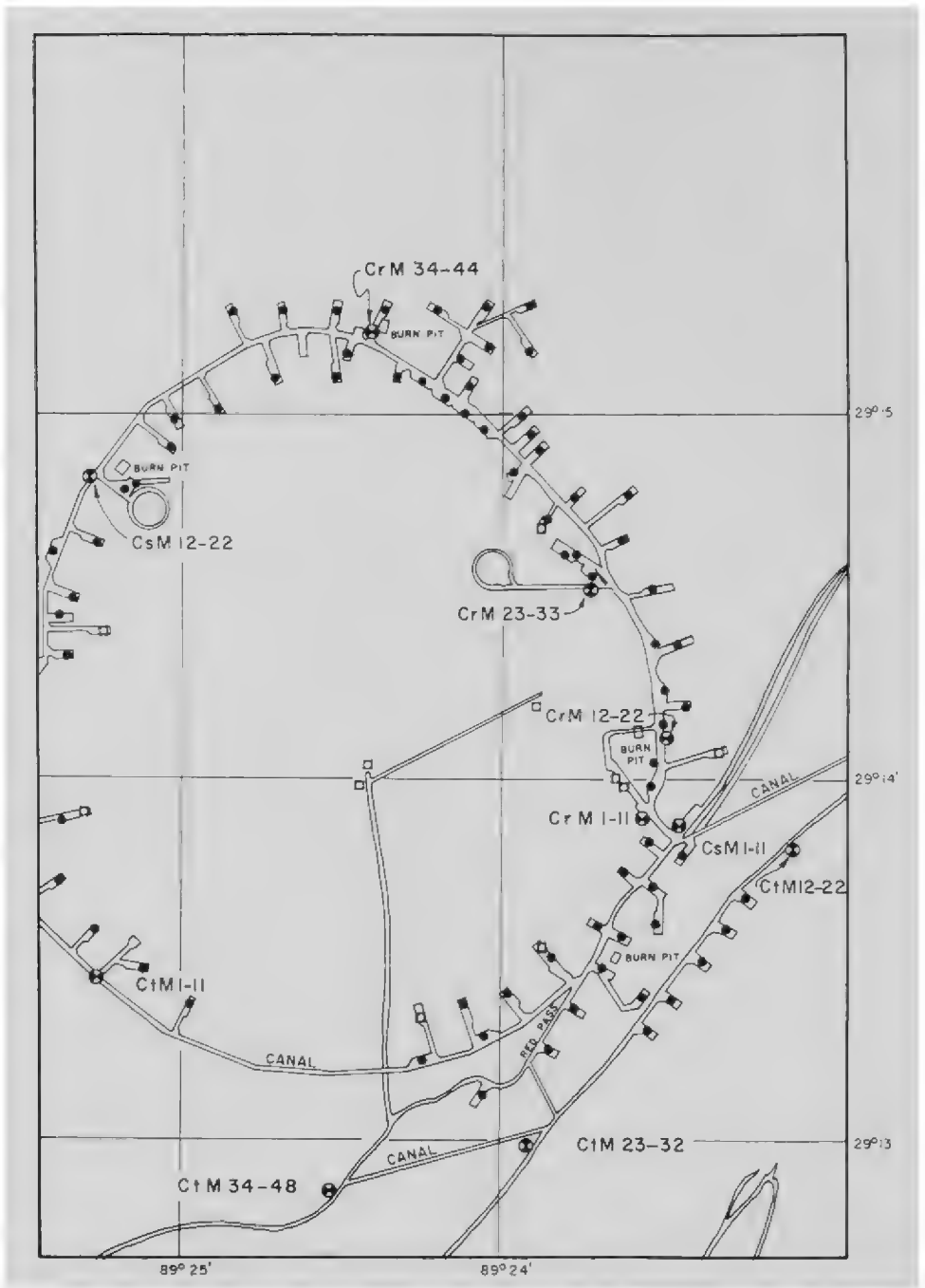

FIG. 28. Canals of the Venice oil field and locations of cores taken for "hydrocarbon" analysis in and around this field.

TABLE 22

Unsaponifiable residues of the muds around the bleedwater outlets at the Lake Pelto Oil Field, November 3–16, 1947, October 5, 1948, and March 30, 1950

Series	Location	Unsaponifiable residue, percent				Pentane and heavier percent
		Composite of cores	Depth in core			
			Top	Middle	Bottom	
Ao 1–11	At BW outlet	0.003	0.002	0.002	0.002	0.0000025
Cv 44	200 ft N of BW outlet	.....	.....	.....	.....	0.0000043
Cv 33	200 ft E of BW outlet	.....	.....	.....	.....	0.0000032
Cv 22	200 ft S of BW outlet	.....	.....	.....	.....	0.0000054
Cv 11	200 ft S of BW outlet	.....	.....	.....	.....	0.0000046
N 1–8	1670 ft E of BW outlet	0.016	.....	.....	.....	.....
Cv 66	4000 ft NW of BW outlet	.....	.....	.....	.....	0.0000033
O 1–11	4700 ft NE of BW outlet	0.013	0.006	0.006	0.005	0.0000042
O 12–22	4700 ft NE of BW outlet	0.016	0.008	0.005	0.009	.....
Cv 45–55	3 miles WNW of BW outlet	.....	.....	.....	.....	0.0000032

TABLE 23

Unsaponifiable residues of muds around the bleedwater outlet at the Caillou Island Oil Field, August 5, 1948, July 20, 1949, and March 29, 1950

Series	Location	Unsaponifiable residue, percent				Pentane and heavier percent
		Composite of cores	Depth in core			
			Top	Middle	Bottom	
Cu 1–11	At BW outlet	0.009	0.012	0.008	0.009	0.0000065
Ao 12–22	At BW outlet	0.004	0.011	0.003	0.003	0.0000043
Cu 23–33	200 ft S of BW outlet	0.006	0.017	0.005	0.004	0.0000058
Cu 45–55	200 ft N of BW outlet	0.007	0.009	0.008	0.010	0.0000037
Cu 12–22	200 ft E of BW outlet	0.005	0.011	0.006	0.006	0.0000030
Cu 34–44	200 ft W of BW outlet	0.006	0.012	0.006	0.006	0.0000059
Cu 56–66	¾ mile from BW outlet at outlying well	0.016	0.012	0.015	0.023	0.0000039
Br 12–22	2 miles from BW outlet*	0.014	0.008	0.015	0.007	0.0000028

* Near a small outlying island.

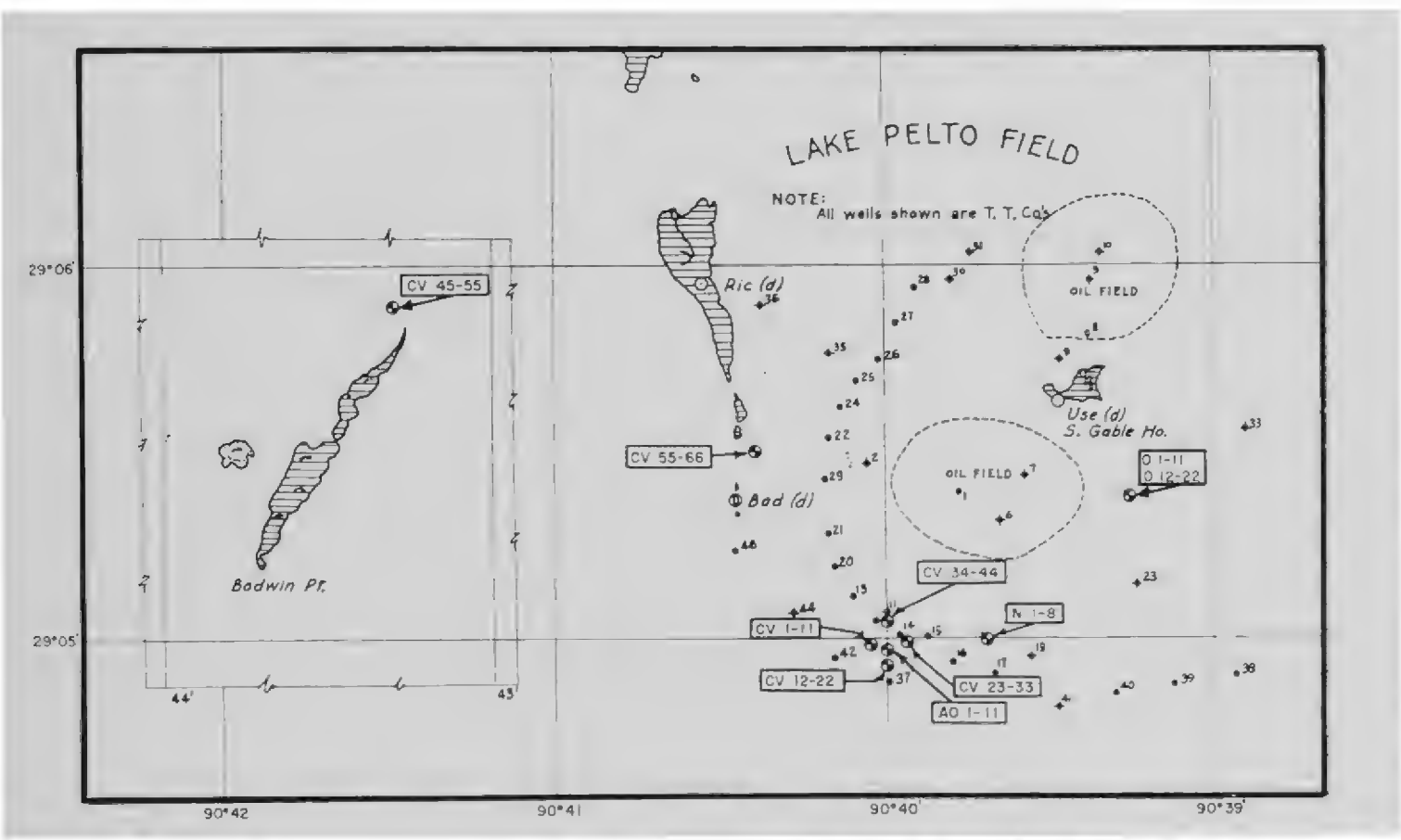

FIG. 29. Locations of cores taken for "hydrocarbon" analysis in the Lake Pelto field.

The Bay Ste. Elaine field is spread over a considerable area of small bays, bayous, and dredged channels in the area just to the west of Terrebonne Bay. Most of the wells are in marsh. Bleedwater production for the entire field in 1947 to 1949 averaged 258 barrels per day. There are two major parts to the field; the one centering around the camp above and below Bay Coon Road and the other lying to the north in the marshes. The cores taken were from the more southerly area. All of the corings were in the field and all were relatively near oil wells, storage tanks, or separators, etc. However, the Au and P series of Table 24 were taken near the bleedwater outlets at the Bay Ste. Elaine camp, and were on the site where an oil loss had occurred. The locations of the mud sam-

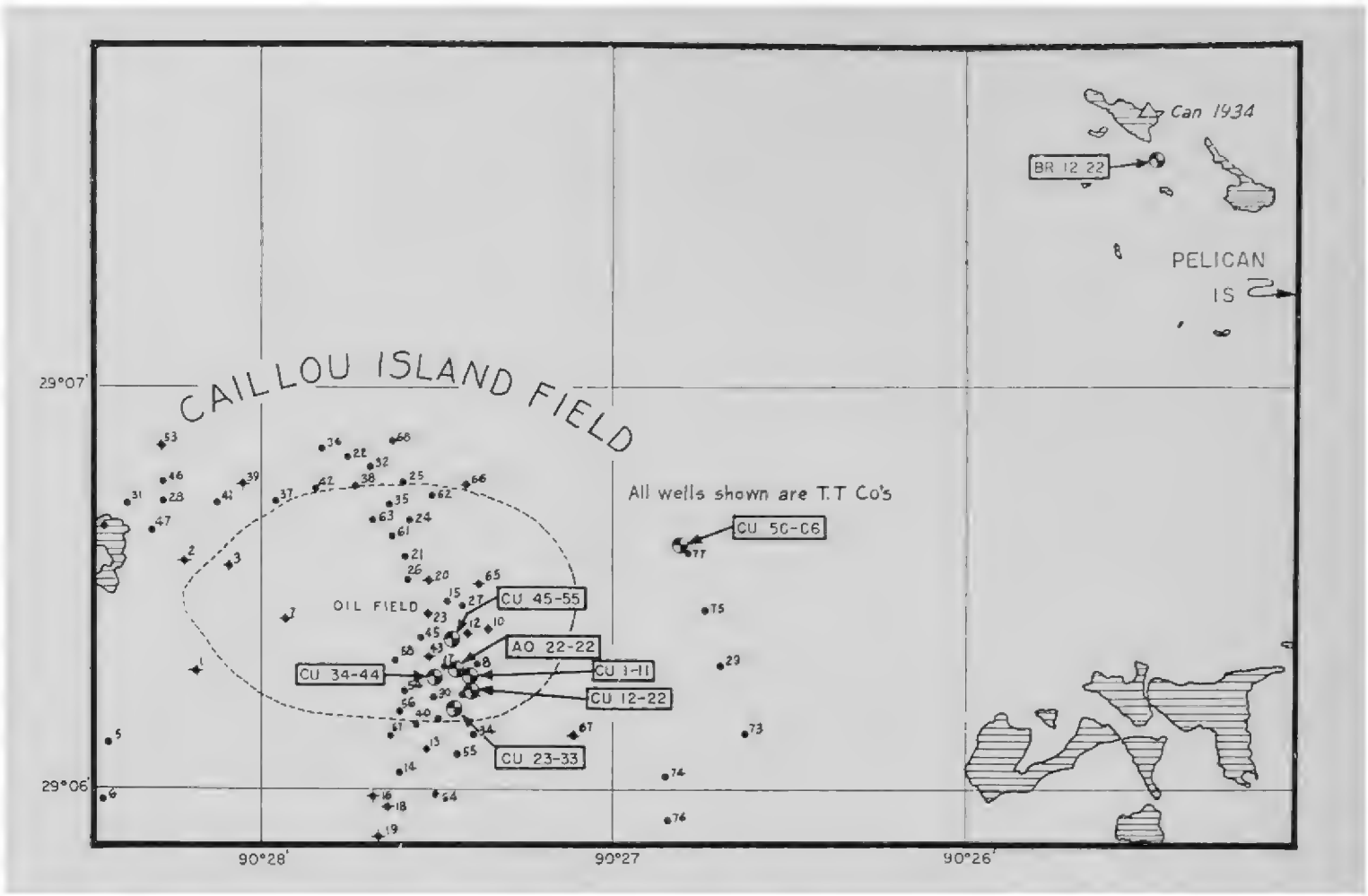

FIG. 30. Locations of cores taken for "hydrocarbons" analysis in the Caillou Island field.

TABLE 24

Unsaponifiable residues of muds in the Bay Ste. Elaine Oil Field, December 22, 1947 to September 1, 1948

Series	Unsaponifiable residue, percent					Pentane and heavier percent
	Composite of cores	Range of values	Depth in core			
			Top	Middle	Bottom	
Au 1	.....	.....	0.018	0.040	0.063	.....
Au 2	.....	.....	0.135	0.023	0.060	.....
Au 3	.....	.....	0.080	0.025	0.310	.....
Au 4	.....	.....	0.021	0.046	0.045	.....
Au 5	.....	.....	0.087	0.024	0.055	.....
Au 6	.....	.....	0.058	0.028	0.047	.....
P 1–12	0.036	0.016 to 0.064	0.054	0.079	0.010	0.0000107
Ad 1–10	0.018	0.016 to 0.035	0.049	0.067	0.005	0.0000071
Ae 1–10	0.029	0.015 to 0.033	0.040	0.010	0.006	0.0000041
Ae 12–22	0.026	0.015 to 0.035	0.035	0.054	0.003	0.0000071
Ae 23–32	0.030	0.013 to 0.036	0.005	0.044	0.003	0.0000065
Ae 34–43	0.034	0.005 to 0.037	0.064	0.195	0.005	0.0000064
Aa 1–10	0.011	0.009 to 0.015	0.055	0.008	0.009	0.0000021
Aj 23–32	0.015	0.010 to 0.015	0.014	0.012	0.007	0.0000158
Aj 12–22	0.010	0.004 to 0.014	0.010	0.009	0.004	0.0000185
Aj 1–10	0.006	0.005 to 0.009	0.008	0.010	0.010	0.0000225
Aj 34–44	0.017	0.008 to 0.017	0.015	0.013	0.004	.....
T 13–22	0.016	0.007 to 0.036	.....	0.043	0.010	0.0000014
T 1–10	0.019	0.015 to 0.065	0.157	0.020	0.041	0.0000053
T 24–33	0.014	0.013 to 0.031	0.031	0.005	0.005	0.0000096
U 1–10	0.043	0.016 to 0.045	0.058	0.014	0.016	0.0000076
U 12–22	0.027	0.009 to 0.052	0.081	0.019	0.007	0.0000030
Y 12–22	0.011	0.005 to 0.044	0.016	0.009	0.016	0.0000019
Y 1–10	0.014	0.007 to 0.024	0.049	0.043	0.009	0.0000015
Z 1–10	0.018	0.012 to 0.021	0.038	0.007	0.007	0.0000026
Z 12–22	0.028	0.014 to 0.042	0.056	0.045	0.013	0.0000053

ples taken in the Bay Ste. Elaine field are shown in Fig. 31. Some of the data contained in Table 24 are also contained in Table 13, which shows values for cores taken in the marshes (see previous discussion).

These data show that there were two local areas of contamination. The first of these was the Bay Ste. Elaine camp itself, where there is a bleedwater discharge, a tank battery, and an oil loading dock, in addition to the wells, and where a local petroleum loss had been sustained. The other contaminated area was in the canals and mud bottom of Bay Coon Road (the Aj series). This series is very interesting in that the detection of petroleum contamination depended on the "pentane and heavier" fraction. The pollution shown here is not considered very significant because the composite values and range of values of the non-volatile fractions were low. The Au series (taken where oil spillage occurred) showed some spots of oil pollution, as indicated by values for "unsaponifiable hydrocarbons", or unsaponifiable residue of extracted material.

Most corings in the Bay Ste. Elaine field showed no pollution, but most did show the effect of peaty layerings in the marshes and along the margins of small bays and bayous. Many oyster experiments were carried out in the Bay Ste. Elaine field (Menzel and Hopkins, 1953). Oysters grow naturally there.

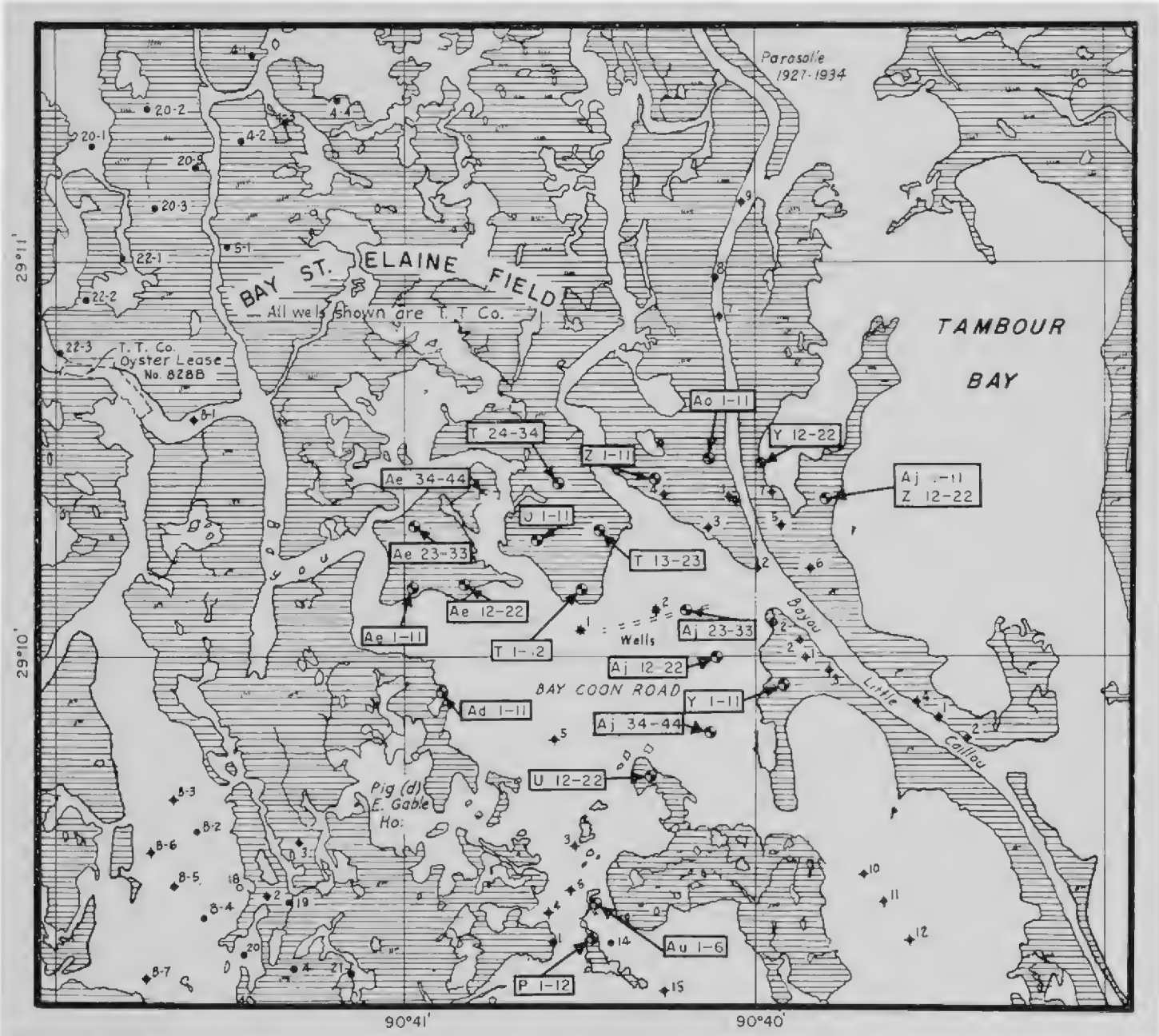

FIG. 31. Locations of cores taken for "hydrocarbon" analysis in the Bay Ste. Elaine field.

Hydrocarbons in the Bottom Muds of Barataria Bay

Data on the concentrations of hydrocarbons in muds around bleedwater outlets have been presented in detail. At some points there was undoubtedly petroleum incorporated in the mud and in some others none was found. It was desirable to compare the values associated with bleedwater outlets with values in the oyster-producing areas. Greater Barataria Bay was selected for test since the claims of oystermen indicated that this was the area of greatest loss of oysters.

Samples taken near the Queen Bess tank battery are included in this table, since bleedwater production here was never more than a trickle and was shut off completely about the time these cores were taken.

About 650 cores, in 68 series, were taken in the Barataria Bay area (Table 25). No

Table 25

Unsaponifiable residues of muds in the Greater Barataria Bay area, August 12, 1947, to February 17, 1950

Location	Unsaponifiable residue, percent				Pentane and heavier percent
	Composite of cores	Depth in core			
		Top	Middle	Bottom	
Hackberry Bay	0.007	0.017	0.009	0.008	0.0000024
Hackberry Bay	0.006	0.017	0.009	0.014	0.0000022
Hackberry Bay	0.007	0.007	0.004	0.005	0.0000027
Hackberry Bay	0.006	0.024	0.018	0.005	0.0000026
Hackberry Bay					0.0000044
Hackberry Bay—south part	0.003	0.005	0.007	0.003	0.0000039
Creole Bay—east entrance	0.017	0.011	0.008	0.043	0.0000020
Creole Bay—east entrance					0.0000035
Barataria Bay—midwest	0.010	0.011	0.030	0.004	0.0000025
Barataria Bay—midwest					0.0000035
Barataria Bay—west side	0.001	0.006	0.003	0.002	0.0000040
Barataria Bay—west side					0.0000037
Barataria—west side	0.008	0.010	0.003	0.005	0.0000099
Barataria—just north of Bassa entrance	0.006	0.017	0.008	0.004	0.0000031
Bassa Bassa Bay					0.0000035
At Bassa Station	0.006	0.009	0.007	0.006	0.0000023
At Bassa Station	0.005	0.004	0.004	0.005	0.0000038
Bassa Pass, south entrance	0.011	0.004	0.016	0.004	0.0000116
W. Barataria, off Mendicant Islands	0.006	0.008	0.006	0.008	0.0000022
W. Barataria, 300 ft north of Queen Bess BW outlet	0.007	0.003	0.000	0.004	0.0000022
W. Barataria, 300 ft west of Queen Bess	0.003	0.011	0.004	0.003	0.0000025
W. Barataria, 300 ft east of Queen Bess	0.004	0.006	0.000	0.003	0.0000032
W. Barataria, 300 ft south of Queen Bess	0.008	0.029	0.003	0.004	0.0000029
Sugar House Bend, south Barataria Bay	0.003	0.003	0.004	0.002	0.0000099
Mouth Bayou Rigaud	0.015	0.010	0.009	0.006	0.0000068
Lab. in Bayou Rigaud	0.017	0.018	0.011	0.007	0.0000019
Bayou Rigaud Sta.	0.009	0.015	0.007	0.004	0.0000051
South Caminada, near Chinatown dock	0.005	0.006	0.004	0.004	0.0000096
Terminus of Southwest La. Canal, east end	0.009	0.028	0.005	0.005	0.0000151
Mouth, Bayou St. Denis	0.013	0.010	0.006	0.023	0.0000022
Mouth, Bayou St. Denis	0.009	0.005	0.005	0.010	0.0000013
North of St. Denis light: near shore, west outlet, Wilkinson Canal	0.009	0.017	0.007	0.003	0.0000214
End of Wilkinson Canal at west outlet	0.012	0.022	0.021	0.026	
Mouth of Wilkinson Canal: east outlet	0.006	0.014	0.008	0.005	0.0000029
West outlet of Wilkinson Bayou	0.008	0.023	0.003	0.004	0.0000113
West of outlet of Wilkinson Canal	0.008	0.016	0.006	0.021	

TABLE 25—Continued

Unsaponifiable residues of muds in the Greater Barataria Bay area, August 12, 1947, to February 17, 1950

Location	Unsaponifiable residue, percent				Pentane and heavier percent
	Composite of cores	Depth in core			
		Top	Middle	Bottom	
St. Mary's light	0.008	0.017	0.010	0.007	0.0000025
St. Mary's Point	0.007	0.017	0.009	0.005	.....
St. Mary's Point	0.007	0.016	0.007	0.010	0.0000014
Mouth, Grand Bayou	0.015	0.020	0.020	0.007	0.0000088
Mouth, Grand Bayou	0.009	0.011	0.005	0.008	0.0000040
Saturday Island	0.012	0.012	0.003	0.004	0.0000010
Bayou Cooper, east side of Bay	0.013	0.012	0.011	0.012	0.0000034
Lower Bay Ronquille	0.003	0.011	0.011	0.006	0.0000045
Lower Bay Ronquille	0.010	0.020	0.011	0.011	0.0000017
Southeast Barataria Bay	0.006	0.006	0.005	0.004	0.0000105
Middle of Bay Long	0.029	0.010	0.010	0.007	0.0000027
Middle of Bay Long	0.009	0.008	0.009	0.017	0.0000036
Middle of Bay Long	0.013	0.006	0.016	0.007	0.0000032
Middle of Bay Long	0.011	0.015	0.004	0.005	0.0000058
Middle of Bay Long	0.010	0.012	0.009	0.010	0.0000040
Middle of Bay Long	0.013	0.015	0.016	0.010	0.0000024
Ronquille Pass (to Grande Ecaille)	....	0.022	0.022	0.005	.....
West Grande Ecaille	0.009	0.009	0.017	0.005	0.0000015
Southeast Grande Ecaille	0.007	.....	0.007	0.005	0.0000035
Southeast Grande Ecaille	0.014	0.024	0.019	0.009	0.0000026
Southeast Grande Ecaille	.....	0.013	0.037	0.007	.....
East Grande Ecaille	0.012	0.045	0.011	0.099	.....
Northeast Grande Ecaille, Mouth, Rattlesnake Bayou	0.028	0.062	0.007	0.099	0.0000031
Mouth, Rattlesnake Bayou	0.022	0.017	0.078	0.012	.....
Big Island, northeast Barataria	0.017	0.023	0.032	0.012	0.0000035
Northeast Barataria: Entry Bay Baptiste	0.008	0.023	0.012	0.004	0.0000045
Northeast Barataria	0.008	0.147	0.003	0.003	0.0000025
Northeast Barataria	0.015	0.037	0.086	0.073	0.0000044
Northeast Barataria	0.013	0.074	0.036	0.006	0.0000042
Northeast Barataria, near Omes Camp	0.010	0.014	0.008	0.014	0.0000024
Northeast Barataria	0.016	0.012	0.054	0.006	0.0000012

place where there was certain contamination of the bottom was found, but pollution was suspected at several isolated spots. The first of these was in the end of the Southwest Canal, where the "pentane and heavier" was high. At that point boat and barge traffic is always heavy. At the south pass to Bassa Bassa Bay, the "pentane and heavier" fraction was possibly a little high. Again, at the end of the west outlet of Wilkinson Canal, the "pentane and heavier" was definitely high, as shown by two core series. It is probable that these local concentrations at the ends of canals with heavy boat traffic are due to the relatively faster currents in the channels. Oil from boats and bilge is adsorbed on silt which is deposited when the current rate drops on entering a bay.

With the exception of the three places to which attention is called, no other part of the Barataria Bay area showed "hydrocarbon" in muds in concentrations large enough to indicate that pollution exists. The values of "hydrocarbons" found in muds of this area, where oyster losses were claimed to be greater than for all other areas combined, were generally low. The values in Table 25 should be compared with those of control samples in Table 10.

Hydrocarbons in Water

Numerous determinations of hydrocarbons in bleedwater were reported by Jensen (1950). The amounts of oil contained in bleedwater vary widely depending on efficiency in separation of salt water from the oil. Jensen reported up to 931 ppm of "non-volatile hydrocarbon" in some bleedwaters produced in southern Louisiana, and as low as 3 ppm in others. Those bleedwaters containing high amounts of oil are usually burned in flare pits or disposed of in some manner not involving discharge into natural waters. Of those discharged into open water, the highest "hydrocarbon" content was 226 ppm at the Caillou Island field. Of 60 determinations, involving 23 bleedwaters, and using from one to three methods of extraction of each bleedwater, the mean value was 35 ppm. Only five out of 60 determinations using three methods showed more than 100 ppm of "hydrocarbons" in bleedwaters. Since some methods of extraction (Jensen, 1950) remove substances other than hydrocarbons from the water samples, the actual mean hydrocarbon content must have been less than 35 ppm.

To determine what effect bleedwaters have on the amounts of "hydrocarbons" found in water around the bleedwater discharges, several studies were made. Data on these are contained in Table 26.

This table shows that the bleedwater has almost no effect on the "hydrocarbon content" of natural waters. Even as close as 25 to 50 feet from bleedwater discharge sites, "hydrocarbon" values were less than that of a control site in the eastern St. Bernard Parish marsh, east of the Mississippi River. Other controls showed low levels of

Table 26

Concentrations of "hydrocarbons" in bleedwater and around bleedwater discharge sites at several oil fields as compared with "hydrocarbons" at great distances from bleedwater discharge areas

Location of samples	"Non-volatile HC", Noll method, ppm
Lafitte Oil Field (8–15–47)	
BW discharge site in canal	5.2
Near BW discharge in canal	2.0
Canal 1400 feet from BW discharge	2.6
Canal in oil field	1.4
Mud Lake, about 6 miles below BW outlet	2.6
Mud Lake, 6–7 miles below BW outlet	3.5
Lafitte Oil Field (10–19–47)	
BW outlet at Camp Lafitte	3.6
75 feet from BW outlet	1.8
1400 feet from BW outlet	3.8
Bayou Cutler about 3 miles below BW outlet	2.4
Bayou Cutler, 5 miles below BW outlet	2.0
Bayou St. Denis, 5.5 miles below BW outlet	1.4
Mud Lake, 6–7 miles below BW outlet	1.4
Delta Farms Oil Field (3–25–49)	
Bleedwater, Delta Farms	5.0
25 feet south of BW discharge	4.0
50 feet south of BW discharge	1.6
200 feet south of BW discharge	3.2
500 feet south of BW discharge	2.4
Delta Farms Oil Field (4–27–49)	
Bleedwater, Delta Farms	7.6
25 feet south of BW discharge	0.6
50 feet south of BW discharge	1.0
200 feet south of BW discharge	0.8
500 feet south of BW discharge	1.6
Halfmoon Lake (East of Mississippi River; control)	1.0

"hydrocarbon" everywhere in Louisiana marsh waters. For example, three samples from marsh ponds showed 0.7, 0.8, and 1.25 ppm respectively. Two samples of water from aquarium supply waters at the Grand Isle Laboratory showed 0.8 and 3.6 ppm.

Owen (1955) attempted to determine whether ether-soluble compounds of the natural waters of Louisiana oyster-producing bays and bayous differed in high mortality areas as compared with those of low mortality areas. His data show no difference in the concentrations, paralleling his findings when comparing the extracts of muds in the same areas. The amounts reported by Owen were very much less than those found by Jensen. He also reported the results of the analysis of samples collected by Louisiana Coastal Waste inspectors. Some of these analyses showed very high concentrations of oil, but Owen pointed out that these cases of high content resulted from taking samples from seepages from oil lines, waste pits, etc., and when compared with his own random sampling, they proved the great extent of dilution. Some of the samples taken by Coastal Waste inspectors and showing high oil content were from areas far removed from the oyster-producing bays.

Studies of Dilution of Bleedwater

Hewatt (1950) made a series of studies to determine how rapidly bleedwater became dispersed and diluted with distance away from discharge points. He pointed out that the highly concentrated bleedwater brines generally contained salts in concentrations of over 100 ppt. The rate of dilution could therefore be approximated by measuring salinity at the bleedwater discharge and at varying distances away from it. For measuring salinity Hewatt used (1) the usual titration method, (2) complete analysis for salts, and (3) a resistivity or conductivity machine. He studied dilutions in this manner at seven different oil fields, some in brackish water and others in oyster-producing areas.

Hewatt made four separate studies at the Lake Salvador oil field. This field produces bleedwater varying from 107 to 131 ppt salinity. The normal salinity of the water in Lake Salvador varies from about 0.1 to 0.25 ppt. Hewatt was unable to trace the effect of the bleedwater farther than 200 feet. He calculated that the greatest possible concentration of bleedwater which could be in the lake was one part in 1892 of water if only the rainfall which fell on the lake was considered. The actual dilution would be enormously greater. This field produced about 2415 barrels of bleedwater daily in 1949.

At the LaFitte field the bleedwater is emptied into a narrow canal which leads into Dupree Cut, another dredged channel which ultimately connects with Barataria Bay. On January 24, 1947, Hewatt found that the salinity of the bleedwater was 128 ppt. At that time the field was producing about 8949 barrels daily (January, 1947, only). At about 1400 feet below the outlet Hewatt found that the salinity had dropped to 1.49 ppt and at 500 feet to the north and to the south in Dupree Cut, the salinity had dropped to about 0.5 ppt. Normal salinity in that area varies from about 0.3 to 10.0 ppt, depending on local rainfall and tidal conditions. Hewatt repeated these observations on February 8, 1947, with almost identical results. On February 23, 1947, he found that at 24 feet from the bleedwater outlet salinity was reduced to 2.81 ppt.

Hewatt made several other studies at the LaFitte field. All of these showed that dilution was extremely rapid, and all showed that mixing was complete and thorough, stratification occurring only within the canal into which the bleedwater outlet emptied.

His conclusions were confirmed at other oil fields, i.e., that thorough mixing always took place.

Hewatt studied dispersal of bleedwater at the Lake Barre oil field above Terrebonne Bay. Continuous records here have shown that the salinity remained near 20 ppt for most of the time. Oysters grow well in this field attached to pilings and even on the mud beneath the wells. Bleedwater was discharged through two "gun barrels" placed close together (20 feet apart), in the amount of 3590 barrels daily during February, 1947. On February 27, 1947, Hewatt found some stratification of salinity as far out as 60 feet from the point of discharge. Salinity of bleedwater was titrated at 149 ppt. Maximum salinity recorded by him on the bottom within 60 feet was 23.5 ppt when his control area showed 20.5 ppt.

Hewatt repeated these studies in February of 1948 with almost identical results, and made other studies at later dates at this oil field. He found thorough mixing of the bleedwater and rapid dilution at all times.

At the English Bay field he found a slight increase in salinity in the water of a short canal into which bleedwater emptied, but no change in salinity outside of it. Later he failed to find any change in salinity following a slight modification in disposal method. A flare is used at the bleedwater pit at this field. At the Venice field, where a number of flare pits are used, Hewatt found no trace of increased salinity in any of the company canals. At the Leeville field he found salt incorporated in the mud of the bottom, and increased salinity in parts of a small reservoir, but no indication of stratification outside of the reservoir. His final conclusion was that thorough mixing of the bleedwaters with surrounding waters took place within a short distance.

Calculations on Dilution of Pollutants

Owen (1955) made some calculations designed to give some idea of dilutions of crude petroleum and bleedwater, and the amounts necessary to make a "one percent soluble fraction" of oil in the bays. Owen had concluded from experiments that a "one percent solution" was "detrimental" to oysters when made up in the manner used in his studies. See experimental section that follows. According to Owen's calculations 767,490 barrels of crude petroleum would be required in a bay 5 miles wide, 10 miles long, and 5 feet deep. This approximates the size of Barataria Bay proper. Owen also calculated that the amount of bleedwater required to make a one percent solution of "soluble fraction" of oil in such a bay was about 3.8 billion barrels, and that with a production rate of 50,000 barrels of bleedwater daily it would require 210 years to build up to that amount. Owen did not consider flushing of his hypothetical bay, but treated it as a closed body.

Jensen (1950) gathered data on the amounts of bleedwater produced in oil fields of southern Louisiana. He used these figures to make calculations designed to show what were the maximum possible concentrations of bleedwaters and crude oils in Barataria Bay. He pointed out that for various reasons it was impossible to calculate exact concentrations in the bays, but that by making certain conservative assumptions, he could arrive at figures which were certainly higher than the actual values.

The results of Jensen's calculations are tabulated below:

(1) Greatest possible increase in salinity in Barataria Bay due to bleedwater ..... 0.00037 ppt

(2) Greatest possible change in the magnesium-calcium ratio of the seawater in Barataria Bay due to bleedwater 0.00000952

(3) Greatest possible concentration of petroleum hydrocarbons in bay waters due to bleedwater ..... 0.000062 ppm

(4) Assuming that the oil companies lost 5,000,000 barrels of crude oil per year, this would produce a concentration in Barataria Bay waters of ..... 6.25 ppm

(5) Assuming that 5,000,000 barrels of crude petroleum was lost in one year and all of it was incorporated into the top 4 inches of mud on the bottom, the concentration would be ..... 0.85 percent

Both the calculations by Owen and those by Jensen assumed that bleedwater and oil must have been distributed over all of the area of Barataria Bay. Research Foundation studies showed that oyster mortalities occurred in all areas. They were not at the same rates everywhere, but in Barataria Bay tended to increase with distance from the oil operations because most oil operations were in brackish-water areas above the bays and the rate of oyster mortality increased with increasing salinity.

Concentrations of bleedwater and oil in a bay may never be the same over an entire bay. Hewatt's studies showed that dilution of bleedwater was rapid and complete, and the concentrations must decrease by at least the square of the distance away from the point of discharge. Crude oil losses, on the other hand, are unevenly distributed in both space and time, and oil may be concentrated within certain areas due to physical obstruction of movement.

Factors Affecting the Length of Time Crude Petroleum May Be Retained in an Area

It has been assumed by some writers that oil is a relatively indestructible substance, and that accidental losses of oil may be cumulative in the bays, especially in the mud of the bottom. Gowanloch (1936) stated, "oil may come to rest in the substratum (of bays) —where it may continue to produce whatever effects are the result of its interaction with sea waters." Smith (in Galtsoff *et al.*, 1935) stated that, "the effect of oil pollution will last over a long period for the oil is carried to the bottom by suspended mud particles and released from time to time by storms, tonging, or dredging."

Jensen (1950) evaporated a considerable number of crudes for about 5 hours at 100°C and found that a loss of from 29 percent to 93 percent took place. He then "weathered" several crudes to determine whether or not they would evaporate at a slower rate under normal temperatures, and found that they lost from 39 to 43 percent by weight after exposure to air for four weeks. He believed that oil when spread in a thin layer on water, and exposed to wind, would evaporate to the extent of 50 percent.

Flushing of oils from the bays unquestionably removes much more oil. However, crude oil has a tendency to strand on mud flats at low tide, and wind may drive it ashore in the marshes where it may stick on the mud. Part of it, as pointed out by Smith, may be carried to the bottom in deeper water by adhering to silt particles. Research Foundation personnel made a study of the fate of oil thus carried to the bottom.

Bacterial Oxidation of Hydrocarbons

ZoBell (1946) pointed out that certain bacteria may have a role in the formation of petroleum and also that others are hydrocarbon oxidizers and destroy crude oils in marine muds. ZoBell, Grant and Haas (1943) found a number of genera of bacteria and actinomycetes which attack and destroy various hydrocarbon compounds in petroleum, and demonstrated that they are widely distributed in marine muds. It became desirable to determine (1) whether or not such hydrocarbon-destroying microorganisms were present in muds of Louisiana bays and (2) if such microorganisms were present, at what rate Louisiana crudes and compounds of crudes were destroyed by them.

ZoBell (1948, 1949) analyzed 108 samples of muds from Barataria Bay and some other locations for the presence of bacteria capable of oxidizing crude oil. The results are presented in Table 27, which is ZoBell's summary.

Table 27

Summary of results obtained from examination of 108 mud samples from Barataria Bay region for presence and relative abundance of aerobic and anaerobic bacteria which oxidize mineral oil. From ZoBell, 1949

Total number of samples	108	Percent
Number showing aerobic oxidizers in 1 gm inocula	101	93.6
Number showing aerobic oxidizers in 0.1 gram samples	80	74.0
Number showing aerobic oxidizers in 0.01 gram samples	39	36.2
Number showing anaerobic oxidizers in 1.0 gram samples	78	72.3
Number showing anaerobic oxidizers in 0.1 gram samples	36	33.3
Number showing anaerobic oxidizers in 0.01 gram samples	12	11.1

These samples were taken at various locations by J. F. Prokop of the Grand Isle Laboratory during 1947 and 1948 and were incubated at the Scripps Institution of Oceanography. Positive inocula of one gram of mud indicated the presence of at least one oxidizer per gram of mud, those of 0.1 gram indicated a minimum of 10 oxidizers per gram of mud, and those samples positive with inocula of 0.01 gram indicated a minimum of 100 oil oxidizers per gram of mud. Only 36.2 percent of the samples indicated that there were more than 100 oil oxidizers; all of the remainder of the samples showed a relatively low concentration of oil oxidizers. Thus, while the microbes utilizing hydrocarbons were shown to be widely distributed (in fact, present virtually everywhere), the relatively small numbers indicated that petroleum hydrocarbons in most of the muds were either small in amount or absent. ZoBell (1949) demonstrated that all crude oils tested (five) from Louisiana were susceptible to oxidation by bacteria in muds from Louisiana bays, and that "bacterial populations of the order of several billion cells per ml of medium developed in the presence of the crude oil."

Rates of Oxidation of Louisiana Crude Oils

Prokop (1950) demonstrated the destruction of crude petroleum by several different methods. Brown (1950), Brown and Van Horn (1950), and Brown, Van Horn, and Reid (1950) all demonstrated the destruction of crude petroleum and various components of crude petroleum by bacteria from Louisiana muds.

Prokop (1950) and ZoBell (1949) measured the rate at which Louisiana crudes were

decomposed by bacteria. Prokop measured the amount of oxygen consumed in oxidation of crude oil by a population of bacteria of 10^6 in a measured period of time. From these data he calculated the amount of methane destroyed. He chose methane because it requires a greater amount of oxygen than any other of the petroleum hydrocarbons and thus would give the most conservative figure. Table 28 taken from Prokop's report shows the amounts of crude petroleum which may be oxidized per given area, with different populations of bacteria.

TABLE 28

Quantities of hydrocarbon oxidized per unit area of mud bottom for different periods of time, at 28°C (Prokop 1950)

Bacterial population (millions per gram)	Grams/acre/ day	Barrels/acre/ year	Barrels/mile^2/ year
1	16.5	0.044	28.2
10	165.0	0.44	282.0
100	1650.0	4.4	2820.0
1000	16500.0	44.0	28200.0

ZoBell made similar calculations, arriving at a figure about one-half Prokop's, at 27°C, or about 14,000 barrels per square mile with a bacterial population of one billion per gram of mud. For citations of other similar studies the reports by Prokop and ZoBell should be consulted.

Temperatures of 27°C are summer water temperatures in Louisiana bays. Winter conditions would bring about a reduction of the rates of oxidation of crude oil. For 10°C of depression of temperature, the rate would be divided by about 2.3 and a further reduction of 10° would reduce the oxidation rate by 2.3^2. Temperatures of 7°C are rare but 17°C probably is close to mean winter conditions.

SUMMARY OF DATA ON HYDROCARBONS

The data presented in this section indicate that muds from the bottoms where oysters were produced were not generally polluted in the sense that there were detectable amounts of crude petroleum present. The amounts of extractable materials were shown to be generally low except at points where bleedwater was directly introduced into waters or where recent oil spillage was known to have occurred. In most cases these points were far removed from oyster-producing waters, but in some others they were in oyster production areas. In all cases the data showed that extractable "hydrocarbons" were rapidly diluted to a low level, and that the amounts actually found in the bays where oysters were produced were low in comparison with controls. In Barataria Bay specifically, "hydrocarbon" values were lower than elsewhere, although this is the area where most of the mortalities were claimed.

It was indicated that the extractable materials in muds generally are of plant origin. This does not preclude the possibility that at any point there may not have been a small fraction of crude petroleum present, but it does show that in no case could the amounts be significant in effect on oysters. Comparisons of amounts of "hydrocarbons" in "high mortality" and "low mortality" areas show that there was no significant difference. Taking into consideration the fact that petroleum hydrocarbons are thought to be

evolved in such areas as the Louisiana marshes, and that in any inhabited area small additions of oil from boats are unavoidable, it would have been surprising if the extractions had shown no traces of hydrocarbons.

Calculations based on the known production of bleedwater and the volumes of water in the bays show also that the amounts of actual petroleum hydrocarbons, if any were present, had to be extremely minute, and probably beyond detection by any method, away from the immediate vicinity of the discharge sites. These calculations confirmed and supplemented the data from chemical measurements.

Studies on bacterial disintegration of crude oils and petroleum products showed that accumulation of petroleum hydrocarbons in the muds is not a possibility, taking into consideration the areas of the bays, volumes of water, and volumes of losses of crude petroleum. This does not apply to local areas at docks or in the immediate vicinity of bleedwater outlets where continuous large accretions occur.

Nothing has been said about the use of visual, taste, and smell appraisals of pollution. However, to investigators in the field it was clear from the beginning that where oil pollution occurred, it could be seen, smelled, or tasted, and could be stirred from the bottom with a stick. With ten years of experience, it has been observed that only a small part of the total area has been affected by the occasional oil losses which occur, and those affected have recovered quickly. In some large areas of high oyster mortality, notably lower Barataria Bay, not even so much as a slick has been observed.

The "hydrocarbon program" yielded figures for the maximum concentrations of oil and bleedwater which might reasonably be expected to occur on or over Louisiana oyster beds. It was then possible to set up experiments using amounts of bleedwater and crude oil in excess of those maximum figures. If these amounts did not cause more oysters to die in the experimental than in the control groups, then it could logically be assumed that the maximum pollution found in the bays did not cause mortality of oysters.

Experimentation with Oil and Bleedwater

Gowanloch (1936) stated: "It became an early surmise of the writer that crude oil itself was the important damaging agent. . . ." He was referring to operations of the oil industry which he considered to be destructive to oysters. He had made certain field and laboratory studies which he believed showed that crude petroleum was highly toxic, at least to oysters.

The field studies set up by Gowanloch consisted of known numbers of oysters placed in wooden trays on racks in areas which he considered to be polluted, and similar trays of oysters on racks in areas which he considered to be free of pollution. In the paper cited above he stated that there were six sets of these trays, of which two contained control oysters and four contained experimental oysters. According to Gowanloch the experimental oysters of one experiment had 46 percent mortality as against 9 percent in three months in controls. In his published paper he did not give locations of either control or experimental stations, or results of the other experiments.

However, in testimony in the case of Doucet vs. The Texas Company, Gowanloch later gave further data concerning these field experiments. These data are assembled in Table 29.

All of Gowanloch's trays and racks were soon riddled by shipworms. The wooden trays

TABLE 29

Results of Gowanloch's field experiments testing the survival of oysters in areas "polluted with crude petroleum" and in "unpolluted" areas. June 15 to September 21, 1933

Number of the rack	Experimental racks and control racks					
	1	2	3	4	5	6
Location	Within 10 ft. of BW outlet at Lake Barre Oil Field	Near BW outlet at Lake Pelto Oil Field*	Bedding ground of St. Pierre near Lake Barre Field	Bedding ground of Ludwig Doucet near Pelto Field	Bayou Beauregard, Mendicant I. Barataria Bay control	Bayou Beauregard, Mendicant I. Barataria Bay control
Oysters per rack	100	100	100	100	50	50
Oysters recovered at end of study	None	17	None	70	None	43
No. dead of those recovered	.....	None	.....	32	.....	4
Percent dead	.....	0.0	.....	46	.....	9

* "Along side the bleedwater discharge."

and supports were not treated in any way. No provision was made for covers to prevent loss of oysters by wave action.

The data in Table 29 show that Gowanloch completed two experiments, of which one showed mortality greater than that in the single control, while the other showed less than the control. Both of these experiments were in the Lake Pelto oil field where conchs (*Thais*) are numerous, while the control was in an ecological area where conchs are much less numerous. Failure to select more nearly equivalent areas for experimental and control stations was unfortunate. Forty-six percent mortality is not excessive in the area of the Doucet bedding ground in summer (see Tables 30 and 31).

TABLE 30

Mortality of oysters kept in wire cages at various stations in Louisiana. Data are from Owen (1955)

Station	Mortality Mar. 22, 1949 to Oct. 18, 1949 percent
Bayou Pierre, in Louisiana Marsh	56
Quarantine Bay, in seed area, east of Mississippi	42
Grand Bay, east of the Mississippi	47
Sandy Point Bay, near end of Mississippi River	48
Bayou Scofield, lower west Delta	58
Bay Adam, upper west Delta	61
Grande Ecaille, east of Greater Barataria Bay area	68
St. Mary's Point, upper Barataria Bay	40
Bassa Bassa Bay, middle Barataria Bay	35
Sugar House Bend, lower Barataria Bay	72
Lake Felicity, above Timbalier and Terrebonne Bays	40
Sister Lake, west of Terrebonne Bay	29

Gowanloch (1936) also reported on a number of laboratory studies on the effect of crude petroleum, sludge, and bleedwater. These were conducted at the Grand Pass Laboratory of the Department of Wildlife and Fisheries. Gowanloch used glass-sided aquaria, 1 by 1 by 2 feet, which held about 30 liters of water. He used a running water system with a rate of flow of 500 cc per minute.

In experiments on the effect of crude petroleum, Gowanloch passed sea water through a small amount (25 cc, originally) of oil confined on the water surface in an open-ended glass cylinder. The cylinder was held in a vertical position in one end of the aquarium.

TABLE 31

Mortality of oysters in Sea Rac trays at stations outside of oil fields in Louisiana. Texas A&M Research Foundation. The period of observations was 14 months in all cases except at Bayou Bas Bleu, where it was 12 months

Location of the station	Mortality in percent
Stations on west flank of Mississippi River Delta, Placquemines Parish:	
Bay Adam	89
Bayou Cook	77
Bastian Island	81
Bay Pomme D'Or	84
Skipjack Bay	77
Bay Jacque	65
Bay Tambour	56
Stations in Barataria and Terrebonne Bay areas:	
Bayou Wilkinson	29
Bassa Bassa Bay	70
Bayou Rigaud	82
Lower Barataria Bay	82
Bayou Bas Bleu	55

The water ran into the cylinder, passed through the oil, and then passed into the aquarium through the open bottom end. The glass cylinder was baffled with fine glass tubing in an attempt to prevent passage of emulsified oil into the aquarium. Small amounts of crude petroleum were added, at unstated intervals, to the supply in the cylinder. In experiments on the effect of sludge, the aquarium was bottomed with a sludge taken from an oil-field tank. One experiment was conducted using a bleedwater mixture in a ratio of 1000 parts of sea water to 1 of oil field brine.

In his studies on the effect of crude petroleum Gowanloch reported mortalities in experimental tanks of 100 percent in 5 months as against 17 percent in controls. In studies lasting 60 days he reported deaths in two experimental tanks as 32 percent and 94 percent as against 2 percent in controls. In a study lasting only 22 days, deaths of oysters in two experimental tanks were reported as 52 and 84 percent, with 16 percent in controls.

An experiment with bleedwater was negative. In one experiment in which an aquarium was bottomed with sludge, 72 percent died, while in another it was reported that 88 percent died. Control mortalities were reported as 17 and 2 percent respectively.

Using Gowanloch's methods, Texas A & M Research Foundation personnel were unable to confirm results of Gowanloch's tank experiments using crude oil (see Table 32, studies 5a and 5b).

Galtsoff *et al.* (1935) presented results of studies on the effect of crude oil and bleedwater on oysters. In this investigation Prytherch and Smith conducted field studies but no field experimentation in Louisiana, in areas where oil and oysters were produced, but failed to find an explanation of extensive mortalities which occurred in the area. Smith observed three widely isolated cases of oil slicks and oil-in-mud, in none of which was abnormal mortality of oysters present. Prytherch pointed out that the mortalities affected only the larger and older oysters, and that the normal community of organisms associated with oysters was present close to oil wells and bleedwater outlets. Prytherch also conducted a number of laboratory experiments at Beaufort, North Carolina, testing survival of oysters when subjected to crude petroleum. He found that heavy concentrations of crude oil had no effect on survival. Galtsoff and Smith, in the same paper, studied the effect of "water soluble fraction" of oil on pumping rate of oysters. They

TABLE 32

A summary of the laboratory studies of the effect of crude petroleum on oysters. These are all of the type where effect is determined on the basis of comparative number of deaths of oysters in control and experimental aquaria

Dates	Experiment			Control			Remarks
	No. oysters	No. dead	Percent	No. oysters	No. dead	Percent	
Study No. 1							
1–6–48 to 7–5–48*	50	23	46	56	40	71	.....
Study No. 2							
12–15–47 to 6–14–48†	50	18	36	100	34	34	.....
Study No. 3a							
7–17–48 to 2–4–49‡	117	68	58	114	68	60	Exper. oysters treated for 15 days, observed 6½ months
Study No. 3b							
8–9–48 to 2–4–49‡	97	25	26	97	30	31	Exper. oysters treated for 10 days, observed 5 months
Study No. 4							
10–30–48 to 4–29–49§	35	31	89	35	30	86	.....
Study No. 5a							
10–28–48 to 4–27–49¶	50	27	54	50	22	43	.....
Study No. 5b							
10–28–48 to 4–27–49¶	50	23	46	50	29	58	.....

* Study No. 1. A test of the effect of crude petroleum emulsified in the aquarium. Oysters were subjected to continuous but fluctuating amounts of emulsified crude oil for a period of six months. Emulsification was by turbulence produced by a strong jet of water into an aquarium with a floating layer of crude oil on the surface. Reference: Mackin, 1948d.

† Study No. 2. A test of the effect on oysters of a substrate of oil-soaked mud. Experimental oysters were kept on oily mud for six months. There were two control aquaria, one bottomed with mud, the other kept clean. There was no mortality difference in these two controls, so data from the two are combined. Reference: Mackin, 1948e.

‡ Study No. 3. Experimental oysters were treated with very heavy dosages of emulsified crude oil, for 10 to 15 days, in an aquarium in the laboratory. At the end of the treatment period, both experimental and control oysters were removed to the Chene Fleur field station. There they were kept in trays for 5 to 6 months and mortality was recorded. Reference: Mackin, 1949.

§ Study No. 4. Experimental and control oysters were selected from a group of oysters with high incidence of *Dermocystidium marinum*, all with heavy infestations of Cliona, Martesia, and Polydora, all thin, and with a high incidence of abscesses, especially of the adductor muscle. Experimental oysters were subjected to continuous dosages of emulsified crude petroleum, to determine whether or not weak and diseased oysters would die at a faster rate when so subjected to oil. Reference: Mackin, 1950a.

¶ Study No. 5. Gowanloch (1934, 1936) reported that his experiments showed that crude oil in very small quantities destroyed oysters in a very short time. Gowanloch's testimony in the case of Doucet vs. The Texas Co. provided sufficient details that the Research Foundation was able to duplicate his studies. Sea water was passed through a small amount of crude petroleum confined in a perpendicular open-bottomed cylinder, which was placed in one end of the experimental aquarium and baffled with broken capillary tubing to prevent the passage of emulsified crude oil into the aquarium. Reference: Boswell, 1950.

made up their water soluble fraction of oil by violent mixing of two parts of oil with one part of water, thus insuring a considerable emulsification of oil as well as extraction of soluble substances. According to these authors, a five percent solution of this mixture inhibited ciliary action of the gills, a conclusion not borne out by their data.

Galtsoff *et al.* (1935) presented data from two studies which were designed to determine whether or not oil and bleedwater had an effect on glycogen content of oysters. In the first study, oil in a heavy surface layer was applied in three experiments. In a period of one month oysters in all three aquaria showed increased glycogen content, and the last check showed that oysters in the experimental aquaria had a glycogen content higher than that of controls. In the second study different concentrations of bleedwater were applied to oysters, and again in three experimental aquaria the oysters gained glycogen. The samples in all of these studies were very small. In the South, oysters generally tend to gain in glycogen content during the winter.

Galtsoff and Smith (in Galtsoff *et al.*, 1935) also studied the effect of bleedwater on feeding rate of oysters and found that a very slight effect was produced by a concentration of 10 percent, and that there was no evidence of permanent injury by repeated subjection to oil bleedwater. Galtsoff and Koehring (in Galtsoff, *et al.*, 1935) set up several

experiments testing the effect of crude petroleum and bleedwater on growth of a diatom (*Nitzschia closterium*). They found that when oil was held on the bottom of culture flasks, growth of diatoms was stimulated as often as it was inhibited. In other studies they showed that 25 percent extractives of oil stirred in sea water reduced growth while 12 percent had no effect. In experiments in which they dialyzed oil through collodion bags, cultures of *Nitzschia closterium* were inhibited in growth; however, the authors did not know what concentrations they were using. Bleedwater also, by their methods, was shown to exert an inhibiting influence on growth in strong solutions. The experiments by Galtsoff and Koehring testing the effect of oil and bleedwater on growth of this diatom are of doubtful significance because the concentrations of oil or bleedwater used were so high that their results could not possibly be applied to conditions in the field in Louisiana.

In spite of the relatively high thresholds found by Galtsoff *et al.*, Galtsoff concluded that under certain unstated conditions oil "may become a deciding factor" in death of the oyster. Prytherch, who carried out the only survival studies on effect of oil, and made the original investigations of the oil wells and oysters in Louisiana, had opinions in sharp contrast with those of Galtsoff as shown by testimony in the case of Doucet vs. The Texas Company.

Owen (1955), studying the effect of oil on oysters in Louisiana, tried to determine what were the actual levels of pollution on oyster beds where mortalities occurred. With his oil experimentation he tied in an extensive series of field observations.

Owen presented data on a total of twenty-one studies of the effect of crude petroleum on oysters. All of these were survival studies. All but three used a "water soluble fraction" of crude oil; this was prepared by churning oil and water (ratio of 1 to 16) in a barrel with a propeller on a flexible shaft turned by a motor. From these studies Owen concluded that a one percent solution was required for any effect on survival. Owen also experimented with bleedwater in studies of survival but considered his experiments to be defective and did not present the data.

Several studies by Owen measured the effect of bleedwater and "soluble fraction" on rate of heart beat in oysters. He devised a complicated machine to record heart rate of oysters (Owen, Maduell and Ingle, 1949). Graphs in his 1955 report showed that bleedwater in concentration of 10 percent and "water soluble fraction" in concentration of 0.1 percent had no effect on heart beat or on shell movements.

Owen added oil in graded amounts to several mixed multures of diatoms, *Chlorella* and other plankters in battery jars. These cultures were carried through a period of over four months and the plankton content was then dried, weighed, and compared with controls. In every case the oil-treated aquaria produced greater quantities of plankton than did the controls and the amounts were in direct proportion to the amount of oil used. This study therefore gave results directly contrary to Galtsoff's conclusions regarding the effect of oil on plankton.

Owen's final conclusion, based on all of his studies, was that "industrial effluents studied are detrimental to oysters in concentrations above 1.0 percent", but he found also that this concentration was not present in natural waters. He concluded that the cause of the unexplained mortalities of oysters was fungus disease caused by *Dermocystidium marinum*.

Collier (1953) described a field study of the effect of crude petroleum on the growth, setting, and survival of oysters. There were several separate substudies in Collier's series.

He held oysters in floating rafts for more than a year under very heavy concentrations of crude petroleum, which floated above the oysters on the water surface. He showed that the oil actually came in contact with the oysters. Growth and survival of oysters thus kept under oil were as good as for the controls, and setting of new spat in the experimental rafts was better than in the control rafts.

Research Foundation Studies of the Effect of Oil on Oysters

It was believed that experimentation on the effect of oil and oil bleedwater on oysters should be carried out largely in the oil fields where pollution was alleged to originate. This would have the advantage that collateral effects of drilling and oil production, whatever they might be, would be tested, as well as the bleedwater and crude oil itself. Some field studies were also conducted at considerable distances from oil operations. Field studies were of two types, those testing for survival of oysters subjected to oil and bleedwater as compared with survival of controls, and those measuring the effect of oil and bleedwater on setting and growth of oysters. Field and laboratory studies were complementary and were planned and interpreted together.

Laboratory studies were divided into two types, (1) testing for survival of oysters subjected to oil and bleedwater, and (2) measuring the capacity of these substances to modify vital functions of the oysters. The latter is the so-called physiological approach. Physiological studies are difficult to interpret in terms of mortality. A modification of function may be demonstrated by application of an irritant, but this does not show that the modification itself is abnormal. If ciliary action of the gills is depressed as a result of application of crude petroleum, it remains to be demonstrated that the depression is abnormal; depression and even complete cessation of ciliary activity occurs under impact of natural changes, such as a decrease in temperature or reduction of certain organic substances in sea water (Collier, Ray and Magnitzky, 1953). Coupled with survival studies, studies of functional changes may be most enlightening, but by themselves they contribute only part of the information necessary to make a decision.

Laboratory procedures which could not be duplicated under field conditions were avoided as much as possible. Laboratory experimentation must be artificial in some basic matters (*e.g.*, the use of a confining aquarium). This is true in lesser degree for field experimentation. Some sacrifice of normality must always be made to secure increased accuracy of data. For example, the ideal way to make a field study is to make plantings of oysters, duplicating methods followed by oystermen. But the difficulty of measuring mortality and growth of oysters under such conditions increases directly as the plantings are made more normal (Menzel and Hopkins, 1952). Any one method obviously has basic limitations, so it is desirable to use many different methods, on the theory that all contribute to solution of the basic problem. In the present investigation all types of studies were carried out, eliminating in each method as many artificialities as possible.

In addition to experimentation in the field, a number of stations were set up for the primary purpose of measuring rates of mortality under different natural environmental conditions (especially under different salinities and temperatures). These measurements provided a base for guidance in interpreting experimental results. In effect, they provided an over-all system of controls. Their greatest contribution, however, lay in providing basic knowledge of the magnitude of mortality rates in Louisiana oysters and the seasonal changes in those rates.

Normal Rates of Mortality in Louisiana Oysters

Owen (1955) presented data on rates of mortality of oysters in various parts of the Louisiana oyster-producing areas. His data are presented in Table 30.

Some of Owen's stations were not in "high mortality" areas; that is, they were in areas not designated by oystermen as areas of heavy loss. Bayou Pierre, far from the oil-producing areas, was also far from areas where high mortality was claimed, but was shown to have quite high losses of oysters. Sister Lake, at that time considered to be a highly efficient seed producer, had the least mortality, but even there it was quite high. Owen stated that there was a positive correlation between high salinity and high mortality at these stations. All of Owen's oysters were more than a year old and most were market size, as shown by his measurements of shell length.

Owen made bottom plantings in Bastian Bay which were destroyed completely in six months. In lower Barataria Bay at Independence Island Owen estimated mortality (from "box" counts) for about ten months at 76 percent.

Gunter (1955) described heavy mortality at a high salinity station (49.7 percent in 5 months) at Port Aransas, Texas, and compared this station with several others in waters of lower salinity. Predators were excluded from Gunter's trays.

Similar studies were made by Research Foundation personnel: Mackin and Wray (1949, 1950), Mackin, Welch and Kent (1950), Menzel (1950d), and Menzel and Hopkins (1952). Records of mortality in areas at some distance from oil fields are shown in Table 31. Oysters were held in Sea Rac trays in all cases, and counts of oysters were made at various times.

All of the figures in Table 31 represent approximate means for a number of separate studies of oysters of various sizes (but none under one year old). For detailed accounts consult the original studies. It was found that the mortality rates were correlated with salinity and that most of the mortality was concentrated in the summer months (Mackin and Wray, 1949, 1950). This agrees with Owen's findings.

Laboratory and Field Studies of the Effect of Crude Petroleum on Oysters

Seven experiments testing the survival of oysters when treated with crude petroleum were carried out in aquaria in the Grand Isle Laboratory. The data from these are contained in Table 32.

Nineteen field experiments were carried out, most of them in the Barataria Bay area, but several in the Timbalier-Terrebonne area. The results of these studies are reported in Table 33.

Physiological Studies of the Effect of Crude Petroleum on Oysters

Physiological studies of the effect of crude oil or extracts of crude oil on feeding activity of oysters were made by E. J. Lund at the Institute of Marine Science, Port Aransas, Texas, and by G. Robert Lunz and associates at Bears Bluff Laboratories, Wadmalaw Island, S. C. The results of their studies are condensed in Tables 35 to 39. A complete account of the Bears Bluff experiments is given in an unpublished Project 9 report by Lunz (1950). The Port Aransas studies were described in Project 9 reports

TABLE 33

Summary of the results of field studies of the effect of crude petroleum on oysters

Dates	Experiment			Control			Remarks
	No. oysters	No. dead	Percent	No. oysters	No. dead	Percent	
Study No. 1							
8–9–47 to 2–9–48*	545	148	27	1121	287	26	.....
10–8–47 to 3–31–48*	394	38	10	702	62	9	.....
1–15–48 to 7–14–48*	172	41	24	338	83	25	.....
Study No. 2							
4–7–48 to 10–27–48†	779	334	43	780	371	48	.....
4–7–48 to 10–8–48†	762	547	72	584	354	61	.....
6–23–48 to 10–8–48†	356	49	14	406	89	22	.....
6–23–48 to 10–27–48†	308	39	13	406	69	17	.....
Study No. 3							
11–24–48 to 6–29–49‡	412	77	19	419	76	18	.....

Dates	Experiment			Control			Remarks
	Spat per tray	Dead per tray	Percent dead	Spat per tray	Dead per tray	Percent dead	
Study No. 4							
8–9–47 to 2–9–48§	194	27	14	265	54	20	Only "boxes", pairs of shells attached at hinge, were counted as dead spat
10–8–47 to 3–31–48§	367	32	9	268	47	18	.....
1–15–48 to 7–14–48§	288	56	19	206	38	19	.....
Study No. 5							
5–2–48 to 1–25–49¶	518	170	33	537	171	32	Ste. Elaine
5–1–48 to 2–6–49¶	1011	137	14	942	217	23	Barre
5–1–48 to 2–3–49¶	161	19	12	95	14	15	Bas Bleu
Study No. 6							
4–7–48 to 10–27–48‖	546	36	7	320	33	10	.....
4–7–48 to 10–8–48‖	618	28	5	1680	144	9	.....
6–23–48 to 10–8–48‖	374	13	4	100	15	15	.....
6–23–48 to 10–8–48‖	704	30	4	1967	53	3	.....
4–7–48 to 10–27–48‖	389	30	8	205	32	16	.....

* Study No. 1. Oysters, kept in trays, were sprayed at (average) weekly intervals with crude petroleum. An ordinary fly spray gun was used for coating the oysters, the mud adhering to them, and the tray containing them with a thick film of oil. Duration of each study was 6 months. Controls, unsprayed, were kept at the same station, Bassa Bassa Station. References: Mackin, 1948a, 1948b and 1948c.

† Study No. 2. Market-sized oysters in trays were kept under a thick layer of crude petroleum confined in a wooden pen, for six-months periods; controls under similar pens but with no oil. Reference: Mackin, 1948b. (These studies were conducted in the Barataria Bay area).

‡ Study No. 3. Oil was kept in pens, in the bottom of which were trays of market-size oysters so placed that at tropic low tide periods the oil was deposited on the oysters in the trays. Controls equivalent except for oil. One-seventh of the amount of crude petroleum used on the oysters destroyed vegetation in an exactly equal area of marsh. Experiments at the Chene Fleur Station. Duration seven months. Reference: Mackin, 1950a.

§ Study No. 4. A study of the setting and survival of spat on oysters and shells in trays sprayed weekly with crude petroleum. Trays were kept on the bottom and on racks at the Bassa Bassa Station. Control trays were treated like the sprayed trays except for the application of oil. References: Mackin, 1948a, 1948b, and 1948c.

¶ Study No. 5. A study of the attachment and survival of oyster spat on oil-soaked wood faggots, as compared with such faggots untreated. Bundles of sticks were attached in trays, examined at monthly and longer intervals, and set of oysters was reported as number per unit area. These studies were at Bay Ste. Elaine, Lake Barre, and Bayou Bas Bleu. Reference: Menzel, 1949.

‖ Study No. 6. A study of the setting and survival of spat on oysters in trays kept under a heavy layer of crude oil. Oil over experimental trays was confined in large pens; controls were kept under similar pens with no oil. At the Bassa Bassa and Sugar House Stations. Reference: Mackin, 1948d.

by Lund (1949), and later published (Lund, 1957).

To test the effect of the "soluble fraction" of crude oil, Dr. Lund used a preparation made by Dr. C. K. Hancock of the A&M College Department of Chemistry. See Hancock (1949). This solution, obtained by counter-current extraction of crude oil with bay water, had a salinity of 16 ppt. In Dr. Lund's first experiment, ten 14-liter aquaria (two sets of five each) were filled with different concentrations of "soluble fraction" in sea

water (sea water only in the two control aquaria). Ten oysters were placed in each aquarium of one set, while the other set of five was left without oysters. Naturally turbid sea water pumped from Aransas Pass was used, without circulation. Turbidity was measured at one or two hour intervals for 10 hours, using a Fisher electrophotometer and recording percentage light transmission compared with light transmission of distilled water (100%). These tests were repeated several times. The results are shown in Table 34. Light transmission at start was 75–78%.

TABLE 34

Effect of various concentrations of "soluble fraction" of crude oil on clearance of turbid sea water by oysters. Lund (1949)

	Concentration of "soluble fraction", percent				
	1	3	6	12	0
Percent light transmission after 10 hours	99	98	97	97*	99

* After 9 hours (last check made).

Galtsoff (in Galtsoff, *et al.* 1935) had tested the effect of "soluble fraction" of crude oil on the water-pumping rate of oysters, measuring the rate by counting drops of water passed through the oyster. Lund (1949) repeated these experiments with certain improvements on methods (using a toy balloon instead of a rubber sheet, and an automatic electric drop counter). The pumping rate varied from 80 to 180 drops per minute. The concentrations of "soluble fraction" tested, 2.5% and 5%, had no effect on pumping rate, according to Dr. Lund. No other data were reported.

In two experiments Lund (1949) tested the feeding rate of oysters in aquaria under a heavy surface layer of crude oil, compared with controls in aquaria without oil. In the first of these experiments re-circulated sea water was used. Oysters were placed on glass dishes, supported above the bottom, in aquaria 1 and 2, each holding 7 liters of turbid sea water. The water in one aquarium was covered with a layer of crude oil (100 cc). An air lift was used to inject a stream of water into the aquarium a short distance below the surface of the water, in such a manner that it continually washed the bottom of the oil layer. Water passed out through a drain pipe near the bottom and was recirculated. Excreta and rejecta of the oysters were collected in the glass dishes to prevent their adding to turbidity. At hourly intervals samples of water were withdrawn for measurement of light transmission, then returned to the system. Control aquaria 3 and 4, without oysters, one with and one without oil, were managed in the same way. Turbidity, originally measured as 83% light transmission, was reduced to 95% in both oyster aquaria in three hours. A second test was run with 25 cc of a heavy silt suspension added to reduce light transmission to 71–78%. Again the level of approximately 95% transmission was reached in three hours in both oyster aquaria. Data, read from Lund's graphs, are shown in Table 35.

The second study of the effect of a layer of crude oil covering the water was considered by Lund (1949) to be "probably the most sensitive and crucial test to determine if oil extracts affect the clearance function of the oyster". Four 50-liter aquaria were supplied by a constant stream of naturally turbid sea water. Two aquaria, A and C, contained 32 young Louisiana oysters. The other two aquaria, B and D, contained no oysters. One liter of crude oil was placed on top of the water in aquaria C and D. In-flowing water

TABLE 35

Effect of a surface layer of crude oil on rate of clearance of turbid sea water by oysters, using re-circulated water. Lund (1949)

Hours after start	Percent light transmission			
	Experiment		Control	
	Aquar. 2 with oysters	Aquar. 4 no oysters	Aquar. 1 with oysters	Aquar. 3 no oysters
Without added silt				
Zero	82.5	82.5	82.5	83
1	82.5	85.5	85	83.5
2	86.5	87.5	91	87
3	95	87.5	95	86
Second run, with silt added				
Zero	78	73.5	78	71
1	88.5	74	82.5	73
2	95	76	90	74
3	96	79	94	77.5

was directed against the oil layer. Turbidity of in-flowing and out-flowing water was measured every 12 hours, and at the end of 11 days the volumes of accumulated silt in the bottoms of the four aquaria were measured. There was no detectable effect of oil on the rate of clearance of water or the rate at which silt (rejecta and excreta) was accumulated by oysters. One oyster died in control aquarium B. The results are shown in Table 36.

TABLE 36

Effect of a surface layer of crude oil on rate of clearance of turbid sea water by oysters, using an open system. Lund (1949)
Clearance was measured by volume of silt removed from water and deposited on bottom by oysters. Silt included faeces (excreta or ejecta) and pseudo-faeces (rejecta). Duration 11 days

	Experiment		Control	
	Aquar. C with oysters	Aquar. D no oysters	Aquar. A with oysters	Aquar. B no oysters
Volume of silt deposited, cc	103	14	98	13

Lunz (1950) studied the effect of "water soluble fraction" of crude oil on oysters, following closely the methods of Galtsoff (in Galtsoff *et al.*, 1935). He used the drop count method of measurement with improvements. Each oyster was tested before application of the oil and again directly afterward. Following Galtsoff, Lunz set up a method of measurement of effect by the following formula:

$$E = \frac{P_1}{P_2} \times 100, \text{ when}$$

E = effect of treatment

P_1 = average pumping rate in "soluble fraction"

P_2 = average pumping rate in sea water

There were in all 6 separate sets of experiments. Various concentrations of "soluble fraction", as shown in the following table, were applied. A summary of Lunz's results taken from his Table 9 is reproduced here as Tables 37 and 38.

TABLE 37

Studies of the effect of extracts of crude petroleum on oysters. Lunz (1950)

Series no.	No. of studies	Concentration of "soluble fraction" percent	Effect of treatment	
			Range	Average
1	17	10	10–233	92
2	18	20	0–179	95
3	6	35	88–113	100
4	8	50	14–97	72
5	5	80	35–111	70
6	6	100	13–75	40

TABLE 38

Studies of the effect of sea water on oysters conditioned in 10% concentration of "soluble fraction". Oysters were changed from the "soluble fraction" solution to sea water, which was considered the "treatment" (exactly the reverse of the experiments using "soluble fraction" for the "treatment"). Three oysters were used, one in one test and the others in two each. Lunz (1950)

Exper. no.	No. of studies	Effect of treatment*		
		Low	High	Average
5	1	69	156	107
44	2	16	89	59
46	2	74	186	141
All studies	5	46	159	98

* Based on time oysters were pumping, only.

STUDIES ON THE EFFECTS OF OIL FIELD BLEEDWATER ON OYSTERS

Oil field bleedwater is waste brine from oil wells of salt dome fields, after separation from oil and gas. A few laboratory studies of the effect of bleedwater on survival of oysters were carried out at the Grand Isle Laboratory as shown in Table 39. Physiological studies on the effect of bleedwater were conducted by G. Robert Lunz and staff at Bears Bluff Laboratories and by E. J. Lund and assistants at the Institute of Marine Science, along with the experiments described above. By far the greater part of the bleedwater studies were carried out in the field, in and around oil fields of the Terrebonne Bay area. Lake Barre, Caillou Island, Lake Pelto, Bay Ste. Elaine, and Dog Lake

TABLE 39

Summary of laboratory studies of the effect of bleedwater on survival of oysters at Grand Isle

Three experiments were made, using bleedwater from Louisiana oil fields. Aquaria were large wooden (cypress) tanks, about 3 by 7 or 3 by 10 feet. The bleedwater was fed into the aquaria, with the sea water, through a V-shaped baffled trough about 10 feet long. Sea water flow was about 4000 cc per minute and the bleedwater flow was adjusted to provide a concentration of 1‰ or 2.5‰. Adult oysters from Bay Chene Fleur were used. Reference: Mackin, 1950c.

No. of exper.	Dates	Experiment			Control			Concentration ppt
		No. of oysters	No. of dead	Percent dead	No. of oysters	No. of dead	Percent dead	
1	6–16–48 to 10–15–48	50	16	32	50	15	30	1
2	11–4–48 to 5–4–49	40	32	80	40	30	75	2.5
3	11–4–48 to 5–4–49	40	29	72.5	40	36	90	1

oil fields was used in studies to test the effect of bleedwater on setting, survival, and growth of oysters under the actual conditions existing in producing oil fields. The results of all studies on the effect of bleedwater on oysters are presented below.

Physiological Effects of Bleedwater

Studies of the effect of oil bleedwater on oysters were carried out by Lunz (1950) and Lund (1949). The results of their experimentation are presented in this section.

Lunz (1950) used the drop counting method to measure pumping rates of oysters in 1%, 5%, and 10% solutions of oil field bleedwater in sea water, as contrasted with pumping rates of controls in pure sea water. The method used was that of Galtsoff (in Galtsoff *et al.*, 1935) with some improvements. He also used Galtsoff's method of computing effect of treatment, eliminating certain objectionable features. The results of Lunz's studies are presented in Table 40. This table is identical with Table 13 of Lunz (1950).

Table 40

Effect of oil field bleedwater on pumping rate of oysters (Lunz, 1950)

Concentration of BW Percent	No. of oysters	No. of observations (changes from sea water to bleedwater)	Effect of treatment (based on entire periods)			Effect of treatment (based on time pumping)		
			Low	High	Average	Low	High	Average
1	9	9	77	139	103	77	139	103
5	10	10	54	161	96	57	161	99
10	12	13	5	48	22	44	143	73

Lund (1949) used the method of clearance of a naturally turbid medium to test the effect of bleedwater on oysters, and to determine the approximate threshold concentration. He gave no figures, but presented graphs of two experiments showing his results. Lund concluded that the threshold solution of bleedwater was above 3%. This agrees with Lunz's results.

Lund also used fuller's earth suspensions in a similar set of experiments. In these only 3%, 9% and 15% solutions of bleedwater were used. The results were similar to those of studies in which naturally turbid sea water was used.

In another experiment, Lund tested the effect of a 10% solution of bleedwater on pumping rate of oysters. Again he gave no figures but stated that Kymograph records showed no effect of the 10% solution on pumping rate.

Lund also stressed the fact that the effects of all concentrations of bleedwater used (up to 15%) were reversible.

Studies on the Effect of Bleedwater on Mortality of Oysters in the Bay Ste. Elaine Oil Field

The Bay Ste. Elaine oil field camp lies between Bay Ste. Elaine and Bay Coon Road, the two minor bays in the maze of bayous, bays, and low marsh to the west of Terrebonne Bay and north of Lake Pelto. The oil field proper lies mostly to the north of Bay Coon Road. Discharge of bleedwater takes place at two points, one at the L. L. and E. tank battery at the Bay Ste. Elaine camp and the other about three miles north on Crooked

Bayou. Both are in confined waters compared with the Caillou Island and Lake Barre areas, but still in major waterways. The Bay Ste. Elaine operating camp, oil installations, and bleedwater outlets are shown in Fig. 32.

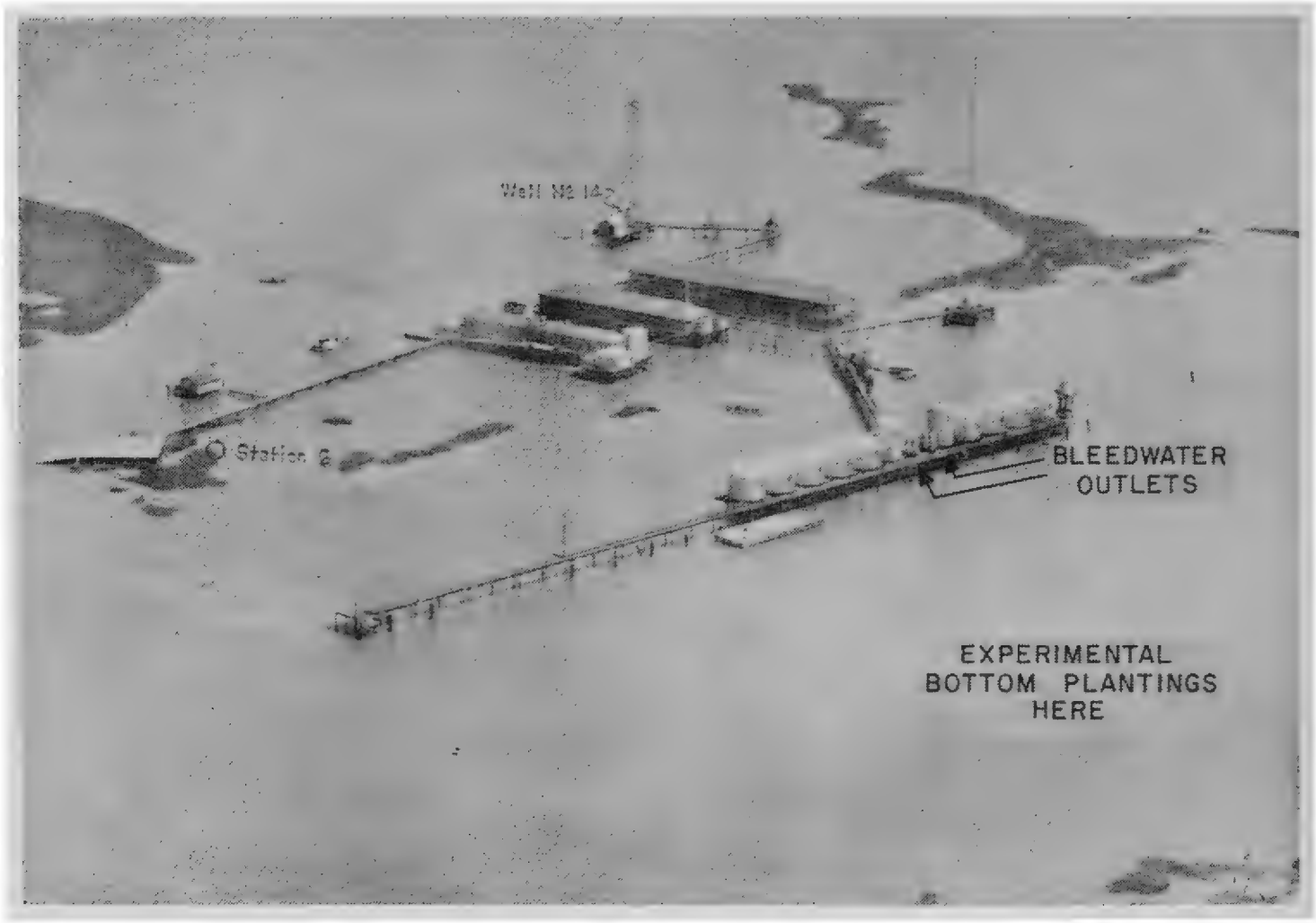

FIG. 32. Aerial photograph of the operating camp of the Bay Ste. Elaine oil field. The tank battery in the foreground released bleedwater at the point indicated. Oyster plantings were made in the channel fronting the tank battery.

The amount of bleedwater discharged in the Bay Ste. Elaine field was quite small (compared with the amounts at Lake Barre and Caillou Island). Both discharge sites together produced only an average of 258 barrels per day from June 1, 1947, to June 1, 1949. The Crooked Bayou site discharged over six times more than the L. L. and E. site.

All of the marsh area around Bay Ste. Elaine produced oysters but the field was in a transition zone of salinity. In the southern part of the area oysters grew naturally only in the intertidal zone. In the northern part of the field and in several small bays just outside the field limits to the north, oysters grew prolifically both below and above the low tide mark. Production in this northern (and inland) area has been good for the entire period from 1947 to 1957, although the Bay Ste. Elaine field has expanded greatly, and since 1950 the Freeport Sulphur Company has established a large mine on Bayou Petite Caillou (closed down at time of going to press).

All of the bays are too shallow for the boats used in oil operations. Thus all well sites are connected by a maze of cut canals, which follow the natural waterways where they exist and dissect the marsh elsewhere. Many of these canals are bordered in places by intertidal mud flats on which grow the usual coon oysters.

Studies of the effect of bleedwater began early in 1947. While the Lake Barre field was made the major site of the field studies on the effect of bleedwater, several experi-

mental investigations were made at Bay Ste. Elaine (Table 41). Reference: Menzel and Hopkins (1953).

TABLE 41

Studies at Bay Ste. Elaine Oil Field testing the effect of bleedwater on survival of oysters.
Menzel and Hopkins, 1953

TABLE 41A

Study No. 1. In October, 1947, two plots of oysters were planted with seed from Bayou Chico. One of these plantings, 200 feet from the L. L. and E. tank battery and bleedwater outlet, was in the "bayou" in the foreground of Fig. 32 which shows the Ste. Elaine camp. The other (control) plot planted was about a mile west in a dredged channel. The ecological factors favored the control plot. The experimental plot had more conchs and stone crabs. Reference: Menzel and Hopkins, 1952, Tables 58, 62, 71, 72, 98, 99; pages 76, 82, 84, 92, 95, 96, 100, 132, 133, and 135.

	Calculated no. of oysters planted	Net yield, sacks per sack planted*	Mortality for a one-year period, percent
Experimental plot 200 ft. from BW outlet	321,000	0.3 to 0.5	82
Control plot at about one mile	354,000	0.8 to 1.2	57

* Net yields are given as of January 18, 1949, when mortality was 86 and 62.5 percent. One year after planting, net yields were 0.4–0.6 and 0.8–1.2 sacks.

TABLE 41B

Study No. 2. A study of the setting and survival of spat. On April 30, 1948, shell bags were placed at two locations, one at about 25 feet from the bleedwater discharge at the Bay Ste. Elaine tank battery, and others at about 350 feet. New bags were put out at intervals. The set was counted and dead spat recorded on shells in all bags in December, 1948. Data on set and mortality are given below. Reference: Menzel and Hopkins, 1953, Table 20, page 42.

		25 ft from discharge		350 ft from discharge	
Period in water	Level above bottom	Spat per shell	Percent dead	Spat per shell	Percent dead
Apr. 30 to Dec.					
2–4	0 ft.	9.2	25	10.6	22
July 2 to Dec.					
2–4	1 ft.	13.4	33	8.0	23
	0 ft.	9.0	31	7.2	23
Sept. 2 to Dec.					
2–4	1 ft.	11.4	26	4.7	52
	0 ft.	15.4	19	1.9	31
All periods and levels, mean	...	11.7	27	6.5	30

TABLE 41C

Study No. 3. The survival of spat at the L. L. and E. bleedwater discharge site was compared with that at a distance of 1600 feet. Five hundred and ten young oysters attached to shells, not more than two months old, were placed in nine hardware cloth baskets and marked with finger nail polish for identification. On June 14, 1947, three baskets were placed about 35 feet from the bleedwater outlet, and six at a distance of 1600 feet to the north-northeast for controls. These latter were moved in August to a location 2600 feet northeast. The experiment ran until April 20, 1949, about 22 months. Reference: Menzel and Hopkins, 1953, Table 23, page 48.

	Distance from BW outlet feet	No. of oysters used	Percent dead†
Experimental oysters	35	180	90
Control oysters	1600 to 2600	330	77

† After 11 months (midway of experimental period) the mortality was 26 percent in experimental oysters and 54 per cent in controls.

TABLE 41D

Study No. 4. Oysters of various sizes and origins, in Sea-Rac trays lined with half-inch mesh hardware cloth, were placed at four stations, two at or near bleedwater outlets and two (control locations) at 2100 and 2160 feet from the nearest bleedwater outlet (at the camp) and two to three miles from the other outlet (in Crooked Bayou). One of the controls was in Bay Coon Road, to the northeast of Bay Ste. Elaine camp (Fig. 31), and the other to the southeast. Experimental trays were placed at various levels, and at various distances from the bleedwater outlets, up to 200 feet away. Because no significant differences in mortality rates were recorded for different levels and distances of the experimental trays, all levels and distances are averaged in the table below. The experiment lasted from August, 1947 to January, 1948. Reference: Menzel and Hopkins, 1953, Table 35, page 67.

	Locations	Distance from BW outlet (feet)	No. oysters used	Percent dead
Experimental oysters	At Ste. Elaine camp	35 to 100	2609	24
	At Crooked Bayou tank battery	100 to 200	602	22
Control oysters	207° from BW at Ste. Elaine	2100	400	22
	340° from BW at Ste. Elaine	2160	1590	20

TABLE 41E

Study No. 5. Oysters from 70 to 100 mm long were placed in 4 Sea-Rac trays, 300 oysters to a tray. Two of these trays were placed 100 feet from the bleedwater outlet at the Ste. Elaine L. L. and E. tank battery and the other two were placed 2160 feet to the north-northwest. The experiment was set up on January 11, 1948 and continued until February 7, 1949. Reference: Menzel and Hopkins, 1953, Table 60, page 96.

	Distance from BW outlet (feet)	No. of oysters used	Percent dead
Experimental oysters	100	600	62
Control oysters	2160	600	47

TABLE 41F

Study No. 6. Oysters about 11 months old were placed in 12 hardware cloth baskets, 50 oysters in each. Four experimental baskets were placed at two levels 25 feet from the bleedwater outlet at the Bay Ste. Elaine L. L. and E. tank battery. Two sets of controls, one examined monthly and the other examined twice in one year, were placed at a distance of 2160 feet to the north-northwest. Each control group consisted of 200 oysters in four baskets. The experiment was begun April 23, 1948, and terminated April 20, 1949, but so many oysters were spilled and lost after October that data are presented here only to October 20, 1948. Reference: Menzel and Hopkins, 1953, Tables 63–66, pages 101–103.

	Distance from BW outlet (feet)	No. of oysters used	Percent dead Oct. 20, 1948
Experimental	25	200	31
Control group 1	2160	200	26 ± 3
Control group 2	2160	200	52 ± 1

TABLE 41G

Study No. 7. Spat up to 5 months old attached to galvanized tin plates were placed in trays. One tray was placed near the bottom 25 feet south of the L. L. and E. bleedwater outlet at the Ste. Elaine camp, and two trays (controls) were placed at a distance of 350 feet to the north. The study began on November 28, 1948. Menzel and Hopkins give data to May 9, 1950 on the experimental oysters, but the controls were lost by accident, one November 2, 1949, and one August 20, 1949. Since the three trays had comparable data to August 20, the mortality data on that date are presented in the table. Reference: Menzel and Hopkins, 1953, Tables 74–76, pages 115–117.

	Distance from BW outlet (feet)	No. of oysters used	Percent dead
Experimental oysters	25 ft S	60	35
Control group 1	350 ft N	48	50
Control group 2	350 ft N	57	44

TABLE 41H

Study No. 8. Oysters about 14 months old were placed in four Sea-Rac trays, 175 to each. Two experimental trays were placed on racks within about 25 feet of the bleedwater outlets at the Ste. Elaine L. L. and E. tank battery; the two controls were placed on racks at a distance of 2160 feet north-northwest. The study began September 18, 1948, and was terminated on April 25, 1949. Reference: Menzel and Hopkins, 1953, Table 108, page 145.

	Distance from BW outlet (feet)	No. of oysters used	Percent dead
Experimental trays	25	350	35
Control trays	2160	350	27

Experiments to Test the Effect on Oysters of Bleedwater Produced in the Lake Barre Oil Field

The Lake Barre oil field of the Texas Company lies in the southern part of the "lake" (see map, Fig. 27). The water in the "lake" has salinity ranging from around 18 to 26 ppt. Around the Lake Barre oil field oysters grow prolifically in the intertidal zones, on pilings and bulkheads and in small reefs on the mud. Subtidal oysters also grow in some areas near the camp, attached to bulkheads. Intertidal oysters are mostly less than eighteen months old.

The field produced about 4000 to 9000 barrels of bleedwater daily, averaging about 6600 per day, from June 1, 1947, to June 1, 1949, the period of bleedwater studies. Thus this field produced the largest amount of bleedwater of any oil field located in oyster-producing waters, and experimentation was more extensive here for that reason. All of the bleedwater is emptied into the water through two "gun barrels" set about 20 feet apart. The experimentation was around these outlets (Figs. 33 and 34). See Table 42.

Table 42

Studies at Lake Barre Oil Field testing the effect of bleedwater on survival of oysters (Menzel and Hopkins, 1951)

Table 42A

Study No. 1. This study was designed to check on the setting and the survival of spat at various distances to the south from the bleedwater outlets and at a control station. Shell bags were placed in the water at two levels and at varying distances on May 1, 1948, and were taken up at intervals of about two months. New shell bags were set in the water when the old ones were taken up for examination. The study was concluded on November 15, 1948. Reference: Menzel and Hopkins, 1951, Tables 4 and 5, pages 16–17.

Dates	Distance from BW outlets (feet)	No. oysters setting per shell	Percent dead
Shell bags 2 feet above bottom:			
May 1, 1948 to July 4, 1948	50	43	12
	85	27	16
	150	35	14
	250	42	21
	2000 (Control)	28	35
July 2, 1948 to Sept. 15, 1948	50	9	19
	85	11	17
	250	13	11
	2000 (Control)	20	18
Sept. 5, 1948 to Sept. 15, 1948	50	2	10
	85	1	41
	150	3	28
	2000 (Control)	1	22
Shell bags on bottom:			
May 1, 1948 to July 5, 1948	50	23	28
	85	30	22
	150	26	17
	250	47	51
	2000 (Control)	22	35
July 2, 1948 to Sept. 5, 1948	50	1	20
	85	9	17
	150	14	13
	250	11	23
	2000 (Control)	15	19
Sept. 5, 1948 to Nov. 15, 1948	50	16	20
	85	11	32
	150	13	19
	250	13	19
	2000 (Control)	6	85

FIG. 33. Aerial photo of the Lake Barre oil field. The point where the bleedwater outlets are located is indicated.

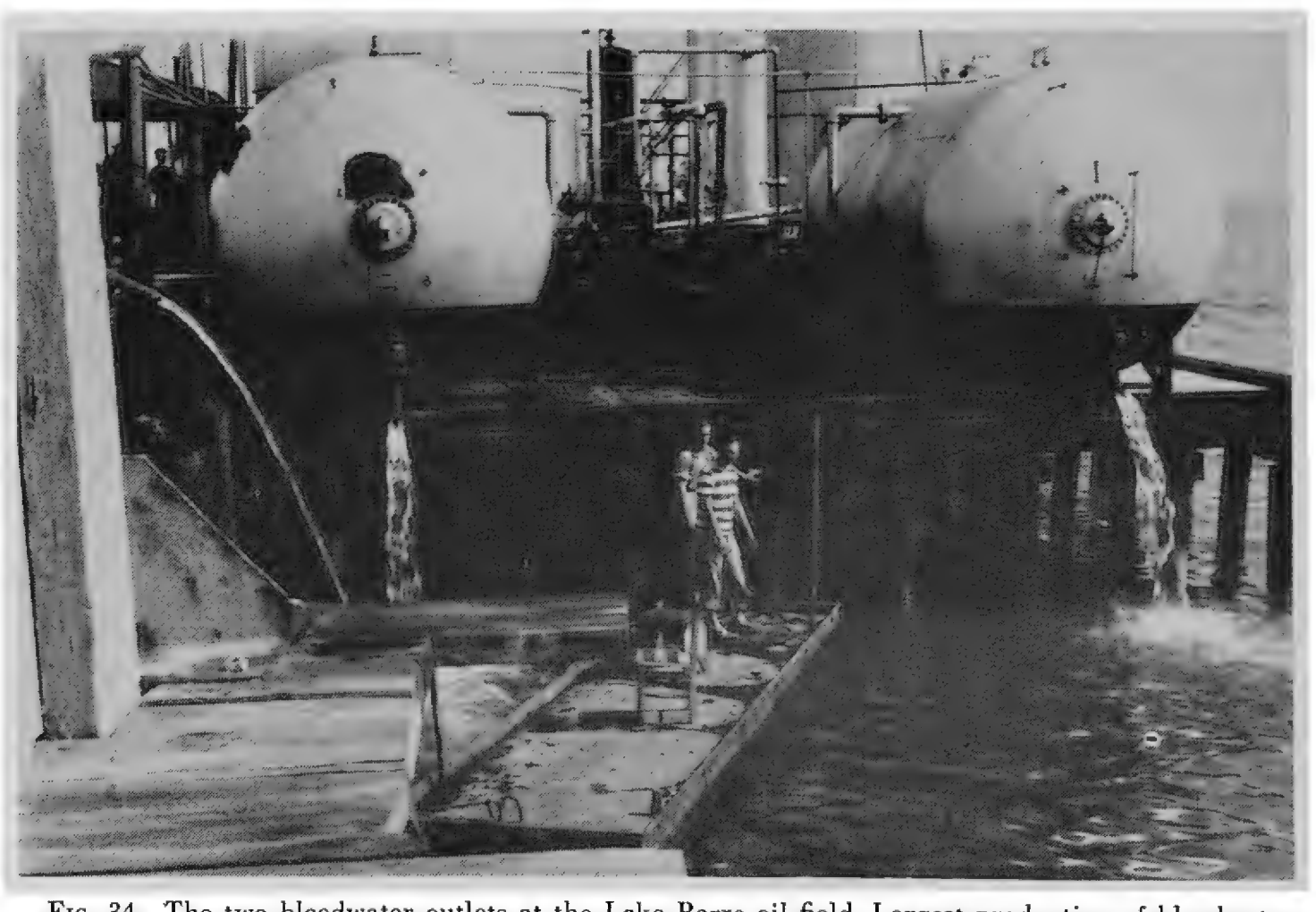

FIG. 34. The two bleedwater outlets at the Lake Barre oil field. Largest production of bleedwater in the south Louisiana area is here.

TABLE 42B

Study No. 2. This study was set up to measure the survival of 2-months-old oysters close to the bleedwater outlets and at a distance of more than ½ mile. Oysters were placed in hardware cloth baskets which were suspended at varying levels above the bottom. They were placed in the water on June 14, 1947. On July 3, 1947 additional young oysters attached to shells were strung on wires in the baskets in such manner that they would not touch the bottom. On July 27, 1947 these young oysters were checked for mortality, as they were again on January 22, 1948. Reference: Menzel and Hopkins, 1953, Table 12, page 26.

Dates	Distance from BW outlets (feet)	Feet above bottom	No. of oysters	Percent dead
July 3, to July 27	80	3	59	24
		1.5	60	12
		0	55	9
	3200	3	60	15
		1.5	60	2
		0	55	13
July 27, 1947 to Jan. 22, 1948	80	3	22	9
		1.5	24	4
		0	24	13
	3200	3	26	4
		1.5	39	13
		0	27	7

TABLE 42C

Study No. 3. Ten-month-old oysters were used in an experiment further testing the effect of Lake Barre bleedwater. Fifty of these young oysters were placed in each of 12 hardware cloth baskets. On April 12, 1948, four of these baskets (experimental) were placed in the water at a distance of 100 feet west of the bleedwater outlets and eight were placed at a control station 2000 feet to the north-northeast. The experiment was terminated on April 21, 1949, a year later. Note that one experimental basket was lost. Reference: Menzel and Hopkins, 1951, Table 14, page 30.

Dates	Distance and direction from BW outlet	Level above bottom (inches)	No. of oysters used	Percent dead in one year
April 12, 1948 to April 21, 1949	100 ft. W	18	50	85
		4	100	71
	2000 ft NNE	18	200	74
		4	200	67

TABLE 42D

Study No. 4. Two hundred oysters approximately 2 months old were placed in each of 15 Sea-Rac trays which were lined with half-inch hardware cloth. On July 16, 1948 these trays were suspended around the bleedwater outlets at various levels, distances, and directions, and at the control station 3500 feet to the south. Since there seemed to be no difference in the two levels used, the different levels at a station are averaged. This study was marred by excessive losses of oysters from some trays with consequent large possibilities for error in the mortality percentage. All trays with such excessive loss were eliminated from this summary. For the full data see the original report (Menzel and Hopkins, 1951), Table 23, page 41. Duration of the study: July 16, 1948 to May 18, 1949.

Distance and direction from BW outlet (feet)	No. of oysters used	Percent dead
25 ft N	392	71
75 ft N	190	7
100 ft S	205	6
150 ft N	204	13
150 ft S	408	14
3200 ft S (Control)	410	20

TABLE 42E

Study No. 5. Oysters of various size classes were placed in Sea-Rac trays on wooden frames at 10 locations in and around the Lake Barre field, as shown in the table below. Some trays were placed near the surface and some near the bottom. There were two control locations, at 3200 feet and 4200 feet. There were excessive losses of trays and oysters due to storm damage. The table presents mean mortality of all size classes for those oysters placed close to the bottom only, since not enough was left of the experimental oysters close to the surface to give significant figures. For the details of both bottom and surface trays see Menzel and Hopkins (1951), Table 32, page 58. The study began August, 1947 and ended in August, 1948.

Direction and distance from BW outlet (feet)	No. of oysters used	Percent dead in one year
40 ft SW	225	99*
80 ft W	710	67
140 ft S	208	48
180 ft W	202	57
240 ft S	206	67
280 ft W	208	62
340 ft S	201	65
500 ft WSW	300	59
3200 ft S (Control)	920	65
4200 ft SE (Control)	300	60

* The original tray of 225 oysters 40 feet southwest of the outlet had 98 percent mortality between August 7 and September 21, 1947. It was replaced by another tray, which also had excessively high mortality. Calculated total mortality was 99.5 percent.

TABLE 42F

Study No. 6. Setting and survival of spat on oysters placed in trays at various distances and directions from the Lake Barre bleedwater outlets in the late summer and fall of 1947 (See Study No. 5). These were counted and checked for total mortality in August, 1948. Reference: Menzel and Hopkins, 1951, Table 44, page 71.

Distance and direction from BW outlet (feet)	No. of trays	Mean spat per tray	Percent mortality
80 ft W	4	102	5
140 ft S	1	500	5
180 ft W	1	193	14
240 ft S	2	170	12
280 ft W	2	401	7
340 ft S	1	190	18
3200 ft S (Control)	5	57	16
4200 ft SE (Control)	1	111	14

TABLE 42G

Study No. 7. Trays of oysters were placed at the bleedwater outlets and at varying distances, directions and levels from the discharge. These oysters were variable in age and size and were random-selected. The study began on October 1, 1947 and ended February 5, 1948. Reference: Menzel and Hopkins, 1951, Table 46, page 81.

Distance and direction from BW outlet (feet)	Distance above bottom (feet)	No. of oysters used	Percent mortality
Between outlets	3	94	23
	1.5	111	95
	0	96	100
20 ft N	3	67	15
	1.5	100	8
	0	109	14
30 ft W	3	66	21
	1.5	78	24
	0	34	65
40 ft N	3	119	14
	1.5	104	11
	0	104	62
40 ft W	3	69	9
	1.5	75	37
	0	75	37
60 ft N	3	105	20
	1.5	106	8
	0	105	61
60 ft W	3	65	12
	1.5	50	14
	0	50	28

TABLE 42H

Study No. 8. About one hundred oysters were placed in each of 42 Sea-Rac trays lined with half-inch mesh hardware cloth. These trays of oysters were placed in various positions around the bleedwater outlet as shown below on April 12, 1948 and taken up April 24, 1949. Oysters were probably about 18 months old on the average, at the initiation of the study. Reference: Menzel and Hopkins, 1951, Table 50, page 87.

Distance and direction from BW outlet (feet)	Level	No. of oysters used	Percent mortality
20 ft W	Top	199	97
	Bottom	199	99
35 ft SW	Top	199	94
	Bottom	197	100
40 ft W	Top	199	98
	Bottom	200	99
50 ft W	Top	201	94
	Bottom	200	93
60 ft S	Bottom	200	90
70 ft W	Bottom	200	90
75 ft S	Bottom	200	93
100 ft W	Top	202	88
	Bottom	202	83
115 ft S	Top	201	79
130 ft W	Bottom	199	93
150 ft W	Top	196	77
	Bottom	202	73
170 ft W	Bottom	198	58
180 ft W	Top	200	70
	Bottom	200	90

TABLE 42I

Study No. 9. Fifteen-month-old oysters were placed in lined Sea-Rac trays. There were 175 oysters per tray. Trays were placed in relation to bleedwater outlets as shown below. The study began October 15, 1947 and terminated May 15, 1949 (7 months later). Reference: Menzel and Hopkins, 1951, Table 54, page 94.

Distance and direction from BW outlet (feet)	Level	No. of oysters used	Percent mortality
25 ft N	Top	169	60
	Bottom	175	91
25 ft SW	Top	173	94
	Bottom	176	100
50 ft S	Top	172	37
	Bottom	176	87
75 ft N	Bottom	174	21
75 ft S	Bottom	175	27
100 ft S	Bottom	175	26
150 ft N	Top	172	26
	Bottom	174	28
3200 ft S (Control)	Top	175	18
	Bottom	172	17
At Bayou Bas Bleu, 5 miles N (Control) †	Top	163	52
	Bottom	169	55

† **Reference: Menzel, 1950c, Table 20, pages 28–29.**

SUMMARY OF THE BLEEDWATER STUDIES AT LAKE BARRE

As a means of summarizing the data on the studies of the effect of bleedwater on oysters at Lake Barre, Table 43, modified from Table 62 in the report by Menzel and Hopkins (1951), is presented. In order to make mortalities comparable, all are reduced to percent per day, and the distances from the BW outlets are combined into segments. Directions are ignored.

TABLE 43

Summary of mortality rates in bleedwater experiments at Lake Barre Oil Field

Distance from BW outlet (feet)	Range of mortality, per day	
	Top trays	Bottom trays
0 to 25	0.18 to 0.25	0.25 to 1.47
35 to 50	0.11 to 0.19	0.27 to 0.49
60 to 75	0.10 to 0.16	0.17 to 0.20
80 to 115	0.20 to 0.21	0.13 to 0.20
140 to 180	0.08 to 0.24	0.09 to 0.19
240 to 500	0.16 to 0.18	0.17 to 0.19
2000 to 4200	0.10 to 0.20	0.11 to 0.18

EXPERIMENTS TESTING THE EFFECT OF BLEEDWATER EFFLUENTS OF THE CAILLOU ISLAND OIL FIELD ON OYSTERS

The Caillou Island oil field is located in the lower part of Terrebonne Bay in a wide expanse of comparatively deep water (maps, Figs 21 and 30), near the large pass into the Gulf. The salinity is high (22 ppt to 30 ppt) in this area. There were three bleedwater outlets, one of which was used in most of the experiments. Production of bleedwater from June 1, 1947 to June 1, 1949, during which period the experiments described below were carried out, was about 2000 to 14,000 barrels daily and averaged about 5000 barrels a day. More than half of this was discharged at the site where nearly all experiments were conducted.

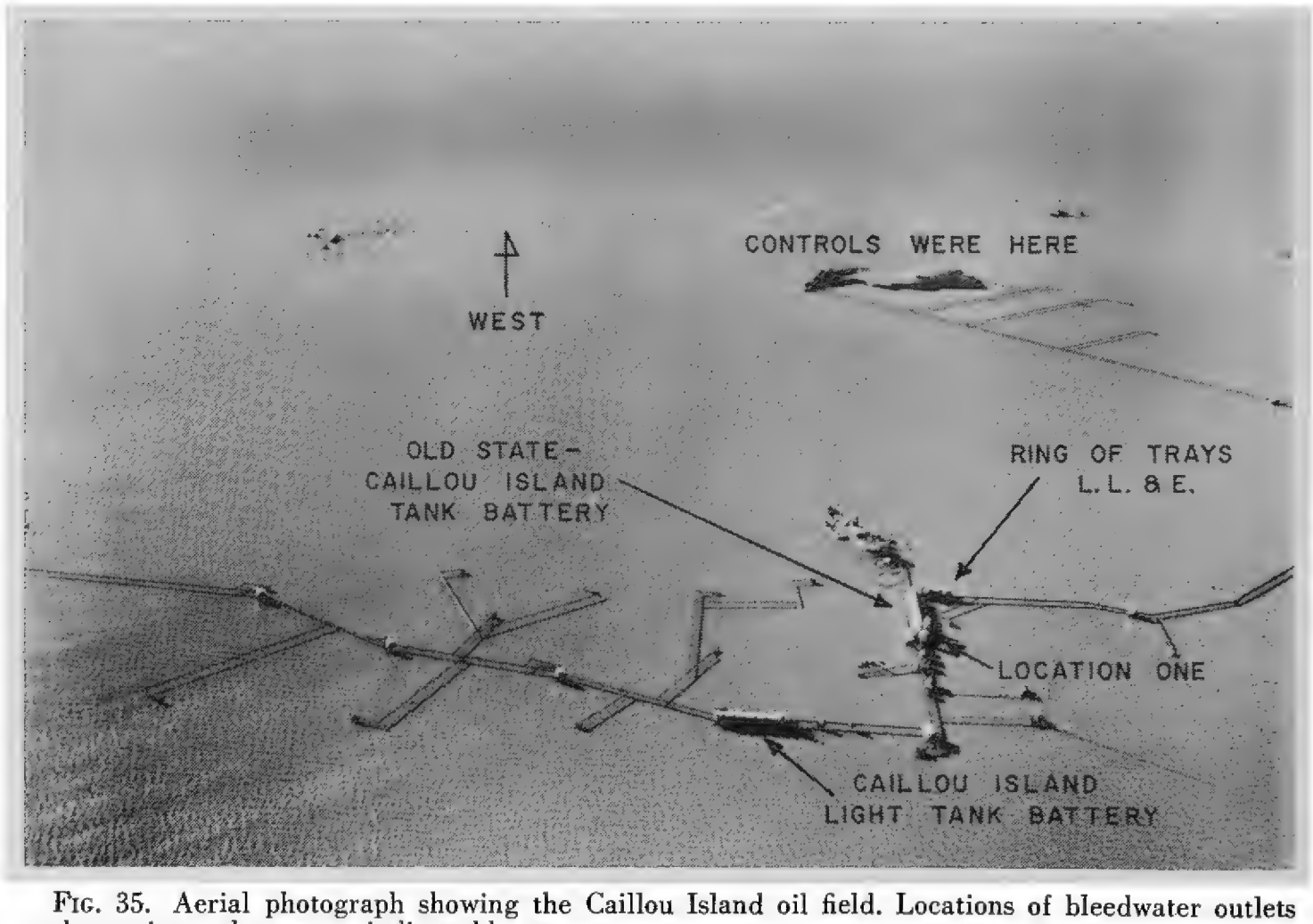

FIG. 35. Aerial photograph showing the Caillou Island oil field. Locations of bleedwater outlets and experimental areas are indicated by arrows.

Fig. 35 shows the general layout of the field, and the expanse of water around it. Oysters grow attached to pilings in the camp, and intertidal reefs are found in coves and bayous of the barrier island to the south. However, no subtidal oysters grow on the bottom and the area long since ceased to be productive of commercial oysters because of the exposed location. Set of spat is good, but unless protected from predators few of these survive for more than a few weeks. Data on experiments carried out in this field are contained in Table 44.

TABLE 44

Studies on the effect of bleedwater on survival of oysters at Caillou Island Oil Field

TABLE 44A

Study No. 1. Three trays of experimental oysters were placed 35 feet from the bleedwater outlet, with 100 oysters in each tray. The oysters were from the Lake Barre oil field. Three trays of control oysters, with 200 to 250 oysters in each tray, were placed 4900 feet from the bleedwater outlet. The control oysters were from Bay Ste. Elaine and were slightly larger than the experimental ones. Reference: Menzel, 1950a, Table 1, pages 5–6.

Dates		No. of oysters	Level of tray*	Percent mortality
Oct. 6, 1947 to Sept. 30, 1948	Experimental	100	Top	71
		100	Middle	60
	Control	250	Top	75
		200	Bottom	73

* The experimental bottom tray and the control middle tray were lost early in the experiment. The experimental middle tray was lost during the last month of the experimental period.

TABLE 44B

Study No. 2. Oysters, in trays containing about 100 each, were placed around a bleedwater outlet at eight points of the compass and exactly 100 feet from the effluent opening. These were compared with two trays 400 feet to the east and two trays 4500 feet to the west. These also held about 100 oysters each. All trays were about 3 feet above the bottom in about 8 feet of water. The experiment began October 17, 1948 and ended April 1, 1949. (The first half of the original experiment, April 4 to October 17, is not reported here because there were no controls until two months after the beginning of the study.) Reference: Menzel, 1950a, Table 3, page 11.

Distance from BW outlet (feet)	No. of oysters	Percent dead
100 ft N	96	19
100 ft NE	99	19
100 ft E	100	13
100 ft SE	100	14
100 ft S	100	16
100 ft SW	100	23
100 ft W	100	17
100 ft NW	99	14
400 ft E	187	15†
4500 ft W	200	10

† There was a possible error of 6% in this tray due to loss of 11 oysters.

TABLE 44C

Study No. 3. An experiment on the setting and survival of spat on trays of oysters near a bleedwater outlet and at a distance. This used trays of oysters in a ring at 16 points of the compass and 100 feet distance from the centrally placed bleedwater outlet. Controls were four trays at 400 feet and four trays at 4500 feet. Depth level was the same everywhere (3 feet above bottom). Period was March 28 (for experimental trays) and May 26 (for controls) to October 17, 1948. The time difference was not significant since set did not occur until May. Reference: Menzel, 1950a, Table 10, page 18.

	16 exp. trays—100 ft		4 cont. trays—400 ft		4 cont. trays—4500 ft	
Period	Spat per tray	Percent dead	Spat per tray	Percent dead	Spat per tray	Percent dead
March 28, 1948 to Oct. 17, 1948	403	4.0	1450	5.6	911	3.7

TABLE 44D

Study No. 4. Dates: September 19, 1948 to April 26, 1949. Three trays with 175 oysters each were placed 100 feet from bleedwater outlets, one north, one northeast and one south. One control tray was placed 4500 feet west of the bleedwater outlet. Oysters were 10 to 16 months old and selected for uniform size. Reference: Menzel, 1950a, Table 12, page 21.

Distance and direction from BW outlet (feet)	No. of oysters	Percent dead
100 ft N	175	16
100 ft NE	175	13
100 ft S	175	30
4500 ft W (Control)	176	35

TABLE 44E

Study No. 5. Dates: May 2, 1948 to November 15, 1948. A check on setting and survival of spat 100 feet and 4500 feet from the bleedwater outlet. Chicken wire bags filled with shells were put in place at the two locations on May 2, and at intervals these bags were taken up and replaced by new ones. Set of oysters and mortality was determined by systematic random sampling of 25 to 30 shells from each bag at the end of each time interval. Bags were placed at two levels; on the bottom and three feet above bottom. Results are reported as average set per shell and percent dead per shell. Reference: Menzel, 1950a, Table 18, page 27.

Distance from BW outlet (feet)	Level above bottom	Total spat per shell	Percent dead
100 ft from BW	3 ft.	15.2	11
	0 ft.	7.0	10
4500 ft from BW	3 ft.	13.9	11
	0 ft.	6.2	11

TABLE 44F

Study No. 6. July 31, 1948 to May 15, 1949. Young oysters two or three months old, all with exactly the same history, were used to test the effect of bleedwater. Four trays each containing 200 of these young oysters were used. Three of these were placed 100 feet north, 100 feet northeast and 100 feet south of the bleedwater outlets; the fourth was placed 4500 feet to the west. Reference: Menzel, 1950a, Table 22, page 35.

Distance and direction from BW outlet (feet)	No. of oysters	Percent dead
100 ft N	201	23
100 ft NE	200	16
100 ft S	200	30
4500 ft W (Control)	200	20

EXPERIMENTATION ON THE EFFECT OF BLEEDWATER ON OYSTERS AT THE DOG LAKE OIL FIELD

The Dog Lake oil field lies in a maze of small "lakes" and bayous to the southeast of Caillou Lake (Sister Lake), and to the northwest of Lake Pelto (Fig. 21). Bleedwater is discharged at two locations, roughly a little more than a mile each from the camp, one to the southwest, and one west. The westerly outlet was used in a single study, and controls were placed at the camp. Fig. 36 shows the locations of the bleedwater outlets and oyster reefs. Oysters grow naturally in this field and all around it. They are particularly abundant around the camp, and are regularly harvested by the oil field workers. Data on experimentation are contained in Table 45.

FIG. 36,A,B. The Dog Lake field camp (36A) and the Dog Lake tank battery, where the bleedwater outlet is located (36B). Experimental oysters were placed at the bleedwater outlet; the controls at the camp, 6,450 feet away.

TABLE 45

Test of the effect of bleedwater on survival of oysters at Dog Lake Oil Field

Study No. 1. Three trays of oysters were placed near the two bleedwater outlets under Unit 12 tank battery, and three control trays at the camp, 6450 feet east of Unit 12 tank battery and 5600 feet from the closest of the three tank batteries. At each location one tray was on bottom, one 18 inches above, and one 36 inches above bottom and a few inches above low tide level. Experimental oysters were from Lake Barre and control oysters from Bay Ste. Elaine. The experiment began October 15, 1947 and continued to October 24, 1948, but two of the control trays were lost before January and the third disappeared before October, 1948, so data are presented only for the periods October 15 to November 16, 1947, and October 15, 1947 to January 17, 1948. Mortality figures are for the trays remaining at the end of the period. Reference: Menzel, 1950c, Table 1, page 5.

Location of trays	No. of oysters	Mortality, percent	
		11–16–47	1–27–48
Tank battery (experimental)	360	3	4
Camp (control)	400	8	18

EXPERIMENTATION ON THE EFFECT OF BLEEDWATER ON OYSTERS AT THE LAKE PELTO OIL FIELD

"Lake" Pelto is a highly saline open-water bay lying to the southwest of Terrebonne Bay and separated from the Gulf of Mexico by a narrow barrier island. There are wide open and deep passes at both ends of the barrier island (Map, Fig. 21). The Lake Pelto oil field of The Texas Company lies near the eastern end of the bay, above Wine Island Pass.

This field produced an average of 1385 barrels of bleedwater daily in the period from June 1, 1947, to June 1, 1949, the period of the experimentation with bleedwater. This amount was discharged into the open water of the bay from four closely clustered outlets (Fig. 37).

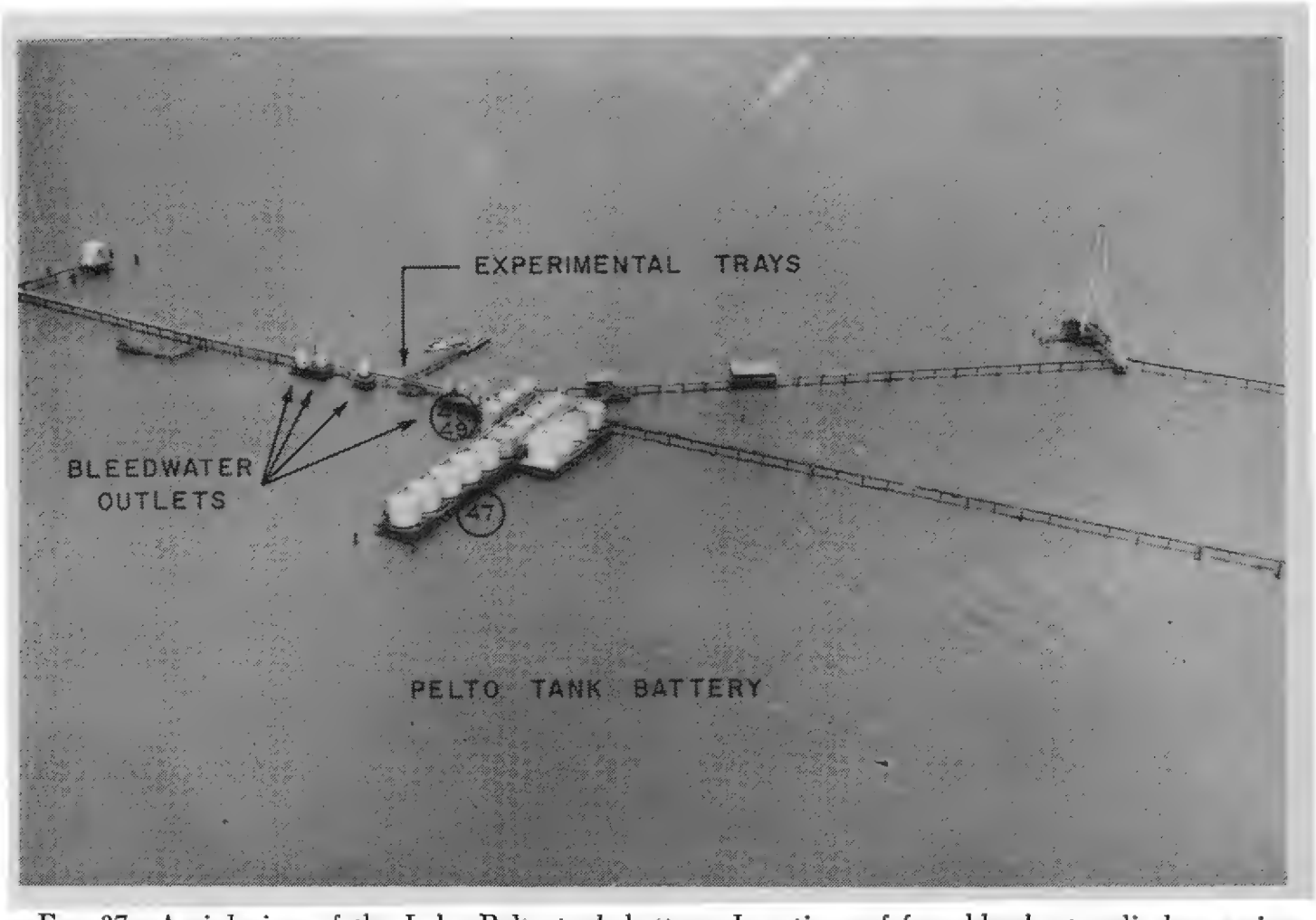

FIG. 37. Aerial view of the Lake Pelto tank battery. Locations of four bleedwater discharge sites are indicated.

Some of the best oysters seen in Louisiana grew attached to timbers in the Lake Pelto oil camp. These were in shaded and sheltered positions. No oysters grow naturally on the bottom below low tide level in Lake Pelto. At certain points there are prolific beds of intertidal coon oysters along the shores of the bay.

Only one study was made testing the effect of bleedwater in the Lake Pelto field (Table 46).

TABLE 46

Study testing the effect of bleedwater on survival of oysters at Lake Pelto Oil Field. Menzel, 1950b

Three trays of oysters of assorted sizes were placed near the four bleedwater discharge sites on October 3, 1947. The nearest discharge site was 35 feet away and another was 125 feet distant on the opposite side. Mortality in these trays was compared with rates in oysters at various distant points in the Louisiana oyster-producing area. The study was terminated on November 29, 1948. Because other studies did not run for comparable periods, the mortality rates are reduced to percent per day for all control stations and the experimental station. The cumulative mortality for the 423-day period was 84 percent (79 percent in the bottom tray and 92 percent in the upper trays).

Reference	Locality	Mortality per day percent
Menzel, 1950d, page 18	Bayou Bas Bleu (Exp. I)	0.17
Menzel, 1950d, page 18	Bayou Bas Bleu (Exp. I)	0.19
Menzel, 1950a, pages 5–6	Caillou I. Camp (Exp. I)	0.21
Menzel, 1950a, pages 5–6	Caillou I. Camp (Exp. I)	0.20
Menzel, 1950b, Table 2	Lake Pelto (4 ft above bottom)	0.22
Menzel, 1950b, Table 2	Lake Pelto (few inches from bottom)	0.19

GROWTH OF OYSTERS SUBJECTED TO CRUDE PETROLEUM

In order to determine whether or not oysters in contact with crude petroleum grow normally, four field studies were carried out at Bassa Bassa Bay in conjunction with survival studies. The data are summarized in Table 47.

TABLE 47

Shell growth of oysters sprayed with oil

TABLE 47A

Study No. 1. Oysters were sprayed with crude petroleum weekly for six months, August 9, 1947, to February 9, 1948. The growth increment of oil-sprayed oysters was compared with that of untreated oysters. There were two groups, one of about 3 to 4-year-old oysters from Bassa Bassa Bay and another of about 2-year-old oysters from Barataria Bay near Saturday Island. The experimental and control oysters were kept in trays at the Bassa Bassa station. Reference: Mackin, 1948a, Table 5, page 8.

Treatment	No. of oysters used	Origin of oysters	Increase in length (mm)
Experiment	169	Bassa Bassa	8.85
Control	381	Bassa Bassa	11.8
Experiment	228	Saturday Island	11.0
Control	453	Saturday Island	9.0

TABLE 47B

Study No. 2. The second study was a duplicate of the first. The period was October 8, 1947 to March 31, 1948, or about six months. The oysters were all about standard seed size (for Louisiana planting), and were from a natural reef in the Lake Barre oil field. Experimental oysters were sprayed with crude petroleum at intervals of about one week for the 6-month period. Reference: Mackin 1948b, Table 3, page 4.

	No. of oysters measured	Increase in length (mm)
Experiment (sprayed with oil)	356	10.7
Control (unsprayed)	640	10.5

TABLE 47C

Study No. 3. Experimental trays of very young spat were sprayed with crude petroleum for a six-month period at weekly intervals. Spat were from three to four months old at beginning of the study on August 9, 1957 and nine to ten months old at time of measurement. Reference: Mackin, 1948a, Table 7, page 10.

	No. of trays	No. of young oysters	No. per tray	Final length (mm)
Experiment	4	269	67	53.2
Control	8	541	68	51.8

TABLE 47D

Study No. 4. Average size attained by oysters attaching and growing in trays sprayed with crude petroleum at one-week intervals, was compared with unsprayed controls. These young oysters were attached to the trays themselves. Since the trays were placed in the water on August 21, 1947, the oldest of the young oysters at the end of the study on March 31, 1948, were about seven months old, and the youngest were about five months old. Reference: Mackin, 1948a, Table 5, page 5.

	No. of trays	No. of oysters attaching	Spat per tray	Final length (mm)
Experiment (sprayed weekly)	2	165	82	36.4
Controls (unsprayed)	4	273	68	36.6

GROWTH OF OYSTERS NEAR BLEEDWATER DISCHARGE SITES

A number of studies made by Menzel and Hopkins in oil fields of Terrebonne Parish were designed to determine whether or not bleedwater could produce stunting of oysters, and if so, at what distances from sources the effect was found. Most of these were carried out in conjunction with the survival studies at the same stations. The data from each oil field are made the subject of a table in which all results of all growth studies at that field are summarized (Tables 48, 49, and 50).

TABLE 48

Growth of oysters exposed to bleedwater at the Bay Ste. Elaine Oil Field

TABLE 48A

Study No. 1. Shell bags were suspended in the water 25 feet from the bleedwater outlets, and the growth of the spat which attached to the shells was compared with growth of spat which attached to shells in a control bag at a distance of 350 feet. Bags were placed in the water at varying times and all were taken up on December 2 or 4, 1948. Reference: Menzel and Hopkins, 1953, Table 20, page 42.

		Average size of spat on December 2–4, 1948 (mm)	
Date placed in water	Position in relation to bottom	Experiment 25 ft from BW outlet	Controls 350 ft from BW
4–30–48	on bottom	16.4 by 13.4	26.8 by 20.4
7–2–48	1 ft above bottom	23.5 by 17.9	29.1 by 23.4
7–2–48	on bottom	16.8 by 14.2	27.7 by 22.1
9–2–48	1 ft above bottom	12.8 by 11.0	14.9 by 13.2
9–2–48	on bottom	16.7 by 13.8	15.7 by 13.8

TABLE 48B

Study No. 2. Young oysters less than 37 days old were placed in wire baskets and measured at irregular intervals for a seven-month period, from June 14, 1947, to January 15, 1948. Two trays were placed 35 feet from each of the bleedwater outlets at Bay Ste. Elaine tank battery. The controls were first kept at a distance of 1600 feet, later changed to 2160 feet. Reference: Menzel and Hopkins, 1953, Tables 24 to 26, pages 45 to 50.

	Size on 6–14–47 (mm)	Size on 1–15–48 (mm)	No. of oysters measured on 1–15–48
Experiment (35 ft from BW)	12.8 by 10.9	72 by 54	59
1600 ft and 2160 ft from BW (Control)	12.1 by 11.0	54 by 42	148

TABLE 48C

Study No. 3. Shell growth and productivity (yield in sacks per sack planted) of oysters at four stations, varying from 25 to 200 feet from bleedwater outlets at the Bay Ste. Elaine L. L. and E. and Crooked Bayou tank batteries, were compared with growth and productivity of controls at two locations, 2100 and 2160 feet from the nearest bleedwater discharge. The experiment ran from August, 1947, to August, 1948. Reference: Menzel and Hopkins, 1953, Tables 30, 49, 51, and 59.

	Experimental locations	Control locations
Number of oysters used	3411	4884
Percent increase in length*	37.8	34.4
Percent increase in width*	34.3	30.4
Percent increase in length × width*	86.5	74.3
Yield, sacks per sack planted†	0.88	0.89

* Average of percentage increases of 19 lots of graded sizes at control locations and 32 lots of graded sizes at experimental locations.

† Average of yields per sack planted calculated for 20 lots of graded sizes at control locations and 31 lots of graded sizes at experimental locations.

TABLE 48D

Study No. 4. Four wire baskets, each containing 50 oysters 10 to 11 months old, were placed within 25 feet of the bleedwater discharge at the L. L. and E. tank battery at Bay Ste. Elaine on April 23, 1948. The control location, 2160 feet to the northwest, also had four baskets with 50 oysters each. Controls and experimental oysters were measured at the beginning of the study and again on April 20, 1949, about one year later. Percent increase in average dimensions and yield in "sacks harvested per sack planted" are shown below. Reference: Menzel and Hopkins (1953), Tables 67, 73, pages 103, 107.

	Percent increase, Mean			Yield, sacks per sack planted
	Length	Width	Thickness	
25 ft from BW discharge	60.3	48.0	70.3	3.0
2160 ft from BW discharge	43.5	33.3	76.3	2.4

TABLE 48E

Study No. 5. Four trays each contained 175 oysters, about 15 months old and ranging in size from 52.5 to 67.5 mm. Two trays (experimental), were placed between the two bleedwater outlets (25 feet from nearest) at the Bay Ste. Elaine L. L. and E. tank battery. The two controls were placed 2160 feet to the northwest. The study began September 18, 1948, and ended on April 25, 1949, about seven months later. Oysters were measured at the beginning and at the end of the study. Yields were computed from growth and mortality data. Reference: Menzel and Hopkins (1953), Tables 113 and 114, pages 148 and 149.

	Percent Increase in 7 months			Yield, sacks per sack planted
	Length	Width	Thickness	
25 ft from BW discharge	38	49	36	1.64
2160 ft from BW discharge	26	33	33	1.52

TABLE 48F

Study No. 6. Bottom plantings were made at two places; one of 1.5 acres was just offshore from the Bay Ste. Elaine L. L. and E. and State tank battery where bleedwater is discharged, the other (control) was in a cut canal approximately one mile west of the Bay Ste. Elaine Camp and comprised 1.4 acres. Natural oysters grew in the latter location, which had a strong scour current and hard bottom. The experimental plot was about average for Louisiana plantings, with a fairly stiff mud bottom. Both areas had heavy boat traffic.

Approximately 600 sacks of seed were planted on each plot, all from the same seed source, in the period October 18–31, 1947. Samples of about three sacks for each plot, one each taken from different boatloads and different parts of each boatload, were measured at time of planting. Samples were taken monthly until January 18, 1949, about 14 months after planting. The oysters in these samples were measured and means computed. Data are presented here comparing the first and last samples from each plot, to show growth in length and width. Reference: Menzel and Hopkins, 1952, Tables 91, 98, and 99, pages 121, 132, and 133.

Location of plot	Length, mm		Width, mm		Yield, Jan. 1949, sacks per sack planted*
	Oct. 1947	Jan. 1949	Oct. 1947	Jan. 1949	
At tank battery (exper. plot)	65.3	96.7	39.6	65.5	0.3 to 0.5
One mile W (control)	67.5	89.6	39.3	56.3	0.8 to 1.2

* The maximum yield obtainable was 1.0 to 1.1 on the experimental plot and 1.1 to 1.3 on the control plot in March and April, 1948.

TABLE 49

Growth of oysters exposed to bleedwater at Lake Barre Oil Field

TABLE 49A

Study No. 1. Wire bags of shells were suspended two feet above the bottom at various distances from the bleedwater outlets. Control bags of shells were placed 2000 feet to the north-northeast. Spat which attached to these shells were measured on November 16, 1948. Only the measurements of the largest 25 percent of the spat are recorded here. These measurements represent approximately two months of growth in 1948. Reference: Menzel and Hopkins, 1951, Table 10, page 23.

Distance from BW outlets (feet)	No. of spat measured	Mean size (mm) of largest 25%	
		Length	Width
50 ft S	256	26.2	20.7
85 ft S	272	26.6	20.6
150 ft S	162	29.0	22.6
250 ft S	201	26.9	20.7
2000 ft NNE	227	26.8	21.9

TABLE 49B

Study No. 2. The second study of growth of oysters exposed to bleedwater at Lake Barre was identical to Study No. 1 execpt that shell bags were on the bottom instead of two feet above. Reference: Menzel and Hopkins, 1951, Table 11, pages 24.

Distance from BW outlets (feet)	No. of spat measured	Mean size (mm) of largest 25%	
		Length	Width
50 ft S	157	13.7	11.5
85 ft S	210	15.8	13.2
150 ft S	266	17.0	13.7
250 ft S	292	19.8	16.9
2000 ft NNE	222	26.0	20.5

TABLE 49C

Study No. 3. Three baskets of young oysters less than one year old were placed 80 feet from the bleedwater discharge at the Lake Barre oil field. Three controls were placed 3200 ft S of the bleedwater discharge. The three experimental baskets held a total of 159 oysters; the three control baskets held 144 oysters. The experiment began on July 27, 1947, when the young oysters were measured for length and width. The study was terminated January 22, 1948, after a period of approximately 6 months. Mortality reduced the measured groups to 84 and 64 oysters respectively. Reference: Menzel and Hopkins, 1951, Table 12, page 26.

	Mean size (mm) July 27, 1947		Mean size (mm) Jan. 22, 1948	
	Length	Width	Length	Width
80 ft from BW discharge	27.8	23.0	53.7	44.2
3200 ft from BW discharge	31.3	25.1	61.5	46.5

TABLE 49D

Study No. 4. Three baskets of young oysters 9 to 11 months old were placed 100 feet W of the bleedwater discharge at Lake Barre, and four others (controls) were placed 2000 feet NNE of the bleedwater discharge. Each basket held 50 oysters. All oysters were measured for length, width, and thickness. The study began on April 12, 1948, and was terminated on April 21, 1949, when all oysters remaining were measured again. Reference: Menzel and Hopkins, 1951, Table 18A, page 33, and Table 21, page 35.

	April 12, 1948				April 22, 1949				
	No. of oysters	Sizes, mm			No. of oysters	Sizes, mm			Yield, sacks per sack planted
		L	W	T		L	W	T	
120 ft from BW outlet	150	54.8	42.5	20.7	36	80.2	56.8	32.9	0.90
2000 ft from BW outlet	200	53.9	42.9	19.1	60	81.0	61.8	33.9	1.15

TABLE 49E

Study No. 5. Oysters about 6 months old were placed at varying distances and directions from the bleedwater outlets at Lake Barre, and their growth was compared with that of oysters of the same age and origin at a distance of 3200 feet from the bleedwater discharges. Because in this study oysters placed 25 to 50 feet from the discharges showed a definite stunting effect, the various locations and levels are reported separately, and the number measured in each case is also given. These oysters had been in position from July 16 to November 7 before the first measurements were made, and the effect of the bleedwater can be seen in the first measurement. Reference: Menzel and Hopkins, 1951, Table 26, page 44, and Table 29, page 47.

Distance and direction from BW outlets (feet)	Number measured 11–7–48	Sizes, in mm			Number measured 5–17–49	Sizes, in mm			Yield, sacks per sack planted
		L	W	T		L	W	T	
Trays 2 feet above bottom									
25 ft N	55	34.5	25.6	13.3	33	42.0	30.6	16.5	0.8
25 ft SW	161	37.8	28.2	15.5	120	37.2	26.9	19.1	4.6
150 ft N	125	39.5	30.1	16.7	115	44.0	33.4	22.4	7.0
150 ft S	190	43.1	32.5	16.6	180	48.5	36.6	22.4	8.6
3200 ft S	181	45.9	35.7	18.1	163	58.7	46.9	25.2	11.3
Trays 4 inches above bottom									
25 ft N	96	33.4	25.8	13.2	81	43.3	34.0	18.3	2.6
25 ft SW	68	31.6	23.9	12.6	12	33.3	24.0	12.1	0.4
50 ft S	161	41.8	30.2	15.7	28	45.0	33.5	20.3	1.2
75 ft N	185	40.8	30.2	15.9	177	53.8	41.4	23.2	9.7
75 ft S	79	43.4	32.3	18.1	75	46.8	34.7	23.5	9.2
100 ft S	195	42.3	32.6	17.1	183	46.1	34.2	22.5	8.0
150 ft N	191	45.3	34.5	17.4	174	59.9	45.8	23.8	11.8
150 ft S	194	46.3	34.9	17.1	156	57.2	42.9	23.0	9.1
3200 ft S	169	46.8	37.6	18.0	142	61.0	49.4	24.3	11.3

TABLE 49F

Study No. 6. Six thousand three hundred oysters of assorted sizes were placed in Sea-Rac trays at various distances and directions from the bleedwater discharge at the Lake Barre oil field. Stations 3200 feet and 4200 feet from the bleedwater outlet were used as control stations. All oysters were measured for length and width of shell at the beginning of the study in early August of 1947 and the survivors were measured again a year later in August, 1948. Increases in size were computed in percent of the original size. Mortalities were high and many oysters were lost in a hurricane and local storms. Reference: Menzel and Hopkins, 1951, Tables 38, 39, pages 62–65.

Distance from BW outlet (feet)	Intial size in mm	No. oysters August 1947	No. oysters measured, August 1948	Percent increase	
				Length	Width
Trays 2½ to 3 feet above bottom					
80 ft W	35–45	100	15	47.2	47.4
3200 ft S	35–45	100	55	85.2	80.5
80 ft W	45–55	135	36	33.7	46.6
240 ft S	45–55	100	29	3.1	23.8
280 ft W	45–55	100	20	31.3	31.3
3200 ft S	45–55	100	24	63.4	61.8
80 ft W	55–60	100	38	28.7	32.1
3200 ft S	55–60	100	48	49.6	47.7
80 ft W	60–70	100	41	15.3	18.8
240 ft S	60–70	100	47	9.4	10.6
280 ft W	60–70	100	27	15.5	15.1
3200 ft S	60–70	100	50	32.6	40.2
80 ft W	70–80	100	37	8.8	14.1
3200 ft S	70–80	100	38	30.5	39.5
80 ft W	90–105	100	26	6.9	21.4
3200 ft S	90–105	100	27	13.6	30.1
Trays 2 to 4 inches above bottom					
80 ft W	35–45	100	35	61.2	58.1
340 ft S	35–45	100	24	65.0	64.4
3200 ft S	35–45	100	34	62.5	68.0
80 ft S	45–55	100	30	40.0	44.1
500 ft SW	45–55	100	35	45.6	48.8
3200 ft S	45–55	102	30	48.1	47.5
4200 ft SE	45–55	100	43	54.7	59.2
80 ft W	55–60	100	38	27.9	35.1
3200 ft S	55–60	100	50	38.7	40.4
80 ft W	60–70	100	33	23.2	23.1

140 ft S	60–70	200	107	37.3	36.9
180 ft W	60–70	100	49	33.7	42.7
240 ft S	60–70	200	65	29.9	33.7
280 ft W	60–70	200	78	28.6	30.1
340 ft S	60–70	100	40	30.8	37.2
500 ft SW	60–70	200	78	33.4	40.2
3200 ft S	60–70	100	43	19.3	39.3
80 ft W	70–80	100	32	18.9	20.7
180 ft W	70–80	100	28	29.0	32.9
500 ft SW	70–80	100	34	21.4	26.4
3200 ft S	70–80	100	36	23.2	15.0
4200 ft SE	70–80	100	37	25.9	30.4
80 ft W	80–90	100	21	13.1	25.6
3200 ft S	80–90	100	36	16.6	33.2
3200 ft S	80–90	200	73	20.7	38.5
80 ft W	90–105	100	17	4.6	2.9
3200 ft S	90–105	100	35	13.0	11.1
3200 ft S	90–105	100	35	9.3	29.1

TABLE 49G

Study No. 7. Oysters 11 to 16 months old and averaging nearly 2½ inches in length were placed at varying distances from the bleedwater discharges at Lake Barre oil field, at two levels: 2 feet above the bottom and about 2 to 4 inches above the bottom. These oysters were measured at the beginning of the study and again at the termination. The study began on October 15, 1948, and ended May 9, 1949, after a period of nearly seven months. Reference: Menzel and Hopkins, 1951, Tables 54, 59, 60, pages 94, 97, 98.

Distance and direction from BW outlet (feet)	No. measured at start of study	No. measured at end of study	Percent increase in L	W	Th	Wt	Vol
Tray 2 ft above bottom							
25 ft N	169	67	11.6	9.0	22.2	46.4	33.0
25 ft SW	173	11	7.2	4.8	11.0	7.7	2.0
50 ft S	172	108	15.0	16.6	20.6	35.6	1.7
150 ft N	172	127	26.2	29.5	28.2	69.1	63.0
3200 ft S	175	144	28.8	28.3	30.6	70.6	68.0
Trays 2 to 4 inches above bottom							
25 ft N	175	15	6.9	—1.0	7.8	9.8	2.3
50 ft S	176	22	5.7	7.4	10.2	51.8	13.9
75 ft N	174	138	21.0	22.6	27.4	54.6	49.8
75 ft S	175	127	22.4	22.1	26.2	62.9	29.7
100 ft S	175	130	17.8	13.8	21.4	47.4	41.6
150 ft N	174	126	26.6	29.9	32.6	84.0	74.3
3200 ft S	172	142	31.9	41.3	31.4	85.1	82.5

TABLE 50

Growth of oysters exposed to bleedwater at Caillou Island Oil Field

TABLE 50A

Study No. 1. Young oysters 10 to 16 months old were placed in four trays, each tray containing 175 oysters selected for size, 52.5 to 67.5 mm in shell length. A sample of 508 oysters was measured for length, width, and thickness on September 19, 1948, and the means were 58.8 by 41.4 by 24.7 mm. Three trays were placed 100 feet from the bleedwater discharge at the Caillou Island field, but in different directions, on September 19, 1948. The control tray was placed 4500 feet west. At the end of the study on April 29, 1949, the oysters were again measured and the growth, in percent of the original size, was computed. Yields in sacks harvested per sack planted were also computed. The data are presented in the table below. Reference: Menzel, 1950a, Tables 16 and 17, page 23.

Distance and direction from BW discharge (feet)	No. oysters measured at end of study	Percent increase: L	W	T	Yield (sacks)
100 ft N	147	34.7	50.2	40.5	2.25
100 ft NE	153	27.7	34.8	38.5	2.11
100 ft S	122	23.5	33.3	34.4	1.46
4500 ft W	113	37.1	44.7	36.8	1.62

TABLE 50B

Study No. 2. Setting and growth of spat were checked in three periods in 1948 by means of chicken wire shell bags. One of the shell bag locations was 100 feet N of the Caillou Island bleedwater discharge site at the Old State-Caillou Island tank battery. The control location was placed 4500 feet W of the BW discharge site. At each location bags were at two levels, on bottom and 3 feet above. Length and width of the spat attaching were measured in millimeters at the end of each of the 3 periods. Reference: Menzel, 1950a, Table 19, page 28.

Period (1948)		100 feet N			4500 feet W		
		Oysters measured	L	W	Oysters measured	L	W
5–2 to 7–5	3 feet above bottom	82	24.8	21.9	100	24.4	19.7
	On bottom	100	24.3	19.2	70	31.0	20.0
7–5 to 9–5	3 feet above bottom	110	10.5	9.0	100	11.2	10.3
	On bottom	107	8.8	7.9	67	10.6	9.3
9–5 to 1–15	3 feet above bottom	99	18.0	14.6	74	19.8	16.2
	On bottom	50	13.5	11.6	78	19.2	15.5

TABLE 50C

Study No. 3. On July 31, 1948, 800 young oysters about 2½ months old were divided into four groups of 200 each, measured for length, width, and thickness of shell, for volume, and for weight. Each group of 200 was placed in one tray. Three of the trays were placed 100 feet from the bleedwater discharge at the Caillou Island field but in different directions. The fourth tray of young oysters was used for a control and was placed 4500 feet west of the bleedwater outlet. All oysters were again measured at the end of about 9½ months on May 15, 1949. Reference: Menzel, 1950a, Table 26, page 38.

Location of station	Percent increase in			Yield sacks
	Length	Width	Thickness	
100 ft N of BW outlet	181.0	195.9	257.5	17.4
100 ft NE of BW outlet	171.3	176.4	238.8	17.0
100 ft S of BW outlet	169.4	174.4	241.3	13.5
4500 ft W of BW outlet	170.2	183.6	237.5	16.0

STUDIES OF ACCIDENTAL LOSSES OF OIL

Several studies were made which involved the measurement of effect on oysters of accidental losses of oil. It is believed that these studies are of primary importance, since the oil was actually observed to come in contact with growing oysters and no artificial laboratory or field equipment was involved. Advantage was taken of the opportunity to study, not only the effect of crude oil losses on oysters, but also the behavior of the oil itself, its distribution, and its duration.

Two studies were made by Menzel (1947, 1948). The first of these involved oysters on beds which were subtidal, and the oil therefore had to be carried down from a surface slick, probably attached to silt particles. About 10 to 20 barrels of oil and sludge were lost as a result of an accident at the Crooked Bayou tank battery of the Texas Company. This is a large natural bayou connecting Bay Sale with Bay Coon Road and with numerous interconnecting waterways, small bays, bayous and canals. The location of the tank battery and the relations of the bayou to other water bodies are shown on the map, Fig. 38. Another oil loss occurred at almost the same time at Well 4–12, about one mile east of this tank battery.

The oil from the tank battery was lost on the night of November 8–9, 1947, and the oil from Well 4–12 was lost October 29, 1947. The effect was noted on an oyster bed one mile to the north on November 9. Oysters there were inspected by Menzel and others on November 11 and found to taste oily. Several testing stations were set up both to the north and to the south of the origin of the tank battery oil, and oysters were tested

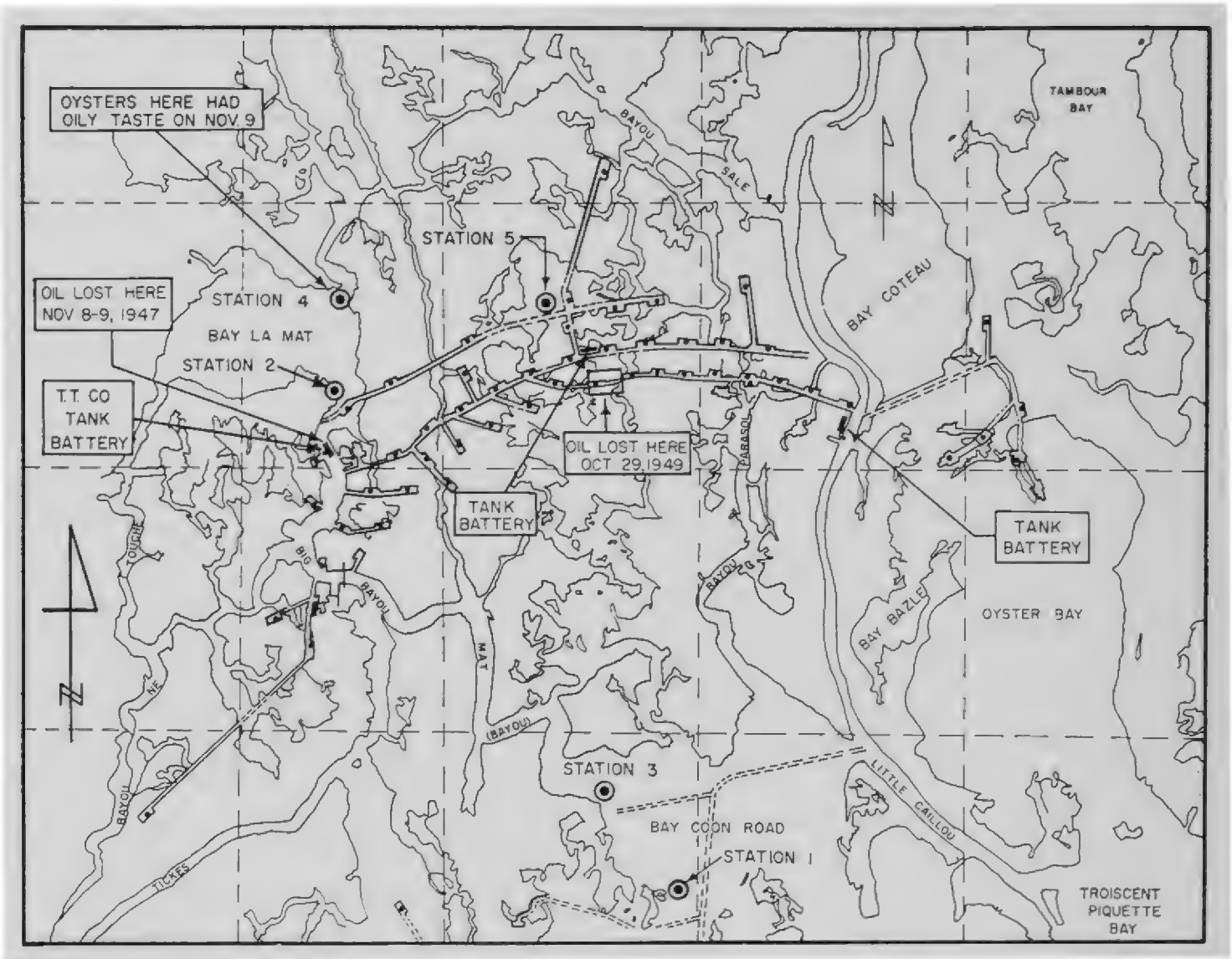

FIG. 38. Site of oil losses on October 8–9, 1947 at Crooked Bayou tank battery, Bay Ste. Elaine oil field, and sampling stations set up to check on the effect of the oil.

for oily taste until it completely disappeared. The results at these stations were as follows:

1. At a station 1000 feet to the north, oysters retained an oily taste to November 21 but had lost it by December 4; thus oily taste disappeared at some time between 14 and 27 days. However, oysters again picked up an oily taste on January 19, and retained it until February 15. Although not continuous, the oily taste in oysters at 1000 feet lasted for more than 3 months.

2. At a station one mile to the north oysters remained oily tasting until November 14, and had no oily taste on November 21 or thereafter. At this station oily taste was retained for 5 to 12 days.

3. At a station two miles to the south, a few oysters were slightly oily in taste to November 14 and the taste had completely disappeared by November 21. Time relations are the same as for the one-mile station but the taste was definitely faint and found in fewer oysters.

4. At a mile and one-third to the south no oily taste was ever observed.

5. At a station about one mile to the east, connected by a cut canal to Crooked Bayou and a short distance from Well 4–12, no oysters were found to have an oily taste. It was evident that water movement was largely along the main bayou and did not move to any great extent through subsidiary waterways. There was no evidence that the Well 4–12 loss caused oily taste in oysters at any time.

This study established that oil lost in a large waterway could be carried in quantity sufficient to make oysters taste oily for at least two miles, and that oysters a mile away

could be made to have strong oily taste for a few days and some oily taste for a period up to 12 days. At 1000 feet, oily taste was recurrent, the recurrence probably resulting from turbulence produced by strong wind or tidal current.

It should be noted also that oil was carried to the bottom immediately. This probably resulted from the fact that a considerable part of the oil from the tank battery was sludge, *i.e.*, weathered oil mixed with dirt. Specific gravity of weathered oil differs but little from that of water, and with the addition of silt, it probably did not float at all.

The oysters affected by this oil loss were watched for the appearance of mortality. The strong oil taste in oysters at 1000 feet and at one mile from the oil loss proved that these oysters had been in direct contact with the crude oil. Samples were taken and "box counts" made. At the first examination it was observed that all "boxes" (empty hinged shells) were old, *i.e.*, they could not have resulted from the oil. It was assumed that if mortality resulted from the contact with oil, the box percentage would increase significantly and that fresh boxes would be observed. This was not the case. Table 51 presents the data from the samples. It is realized that the percentage of boxes cannot be translated into actual rate of mortality.

TABLE 51

Percentage of "boxes" (empty pairs of shells attached at hinge) in samples of oysters tonged at two stations in the Bay Ste. Elaine area after an oil loss, 1947–1948

Date 1947–48	Percentage of boxes	
	At 1000 feet	At one mile
November 11	.....	8.0
November 12	10.4	.....
November 14	.....	12.4
December 4	8.2	12.5
December 14	15.8	11.0
December 28	14.9	8.5
January 19	14.0	11.5
February 9	13.2	.....
February 15	10.8	18.7
March 8	8.8	7.9
April 10	12.1	11.2
May 9	8.5	16.1
June 9	18.9	5.9

TABLE 52

Percentage of "boxes" in random samples of oysters tonged in various areas

Location	Percentage of boxes
Bayou BasBleu	12.8*
Bay Ste. Elaine	21.7†
Bayou Du West	17.5
Northern Barataria	37.0‡
Bastian Bay	35.2§
Chimney Bayou	21.4‖
Bassa Bassa Bay	7.4
Sandy Point Bay	19.7¶
Bay Coquette	21.5
Skipjack Bay	23.7
Saturday Island	3.1
Pickett Lake	22.7**

* Average of 17 samples taken at different periods of the year.
† Mean of 10 groups of box count samples each including 6 to 15 individual samples.
‡ Mean of 6 samples.
§ Mean of 4 samples.
‖ Mean of 7 samples.
¶ Mean of 2 samples.
** Mean of 5 samples.

Glycogen analyses showed that these oysters remained in good condition until spawning took place in the spring, when the usual radical drop occurred.

Box counts have been made from samples of oysters scattered all over the oyster-producing area of Louisiana. In Table 52 below are a few random analyses from various places, for comparison with those of Table 51.

A second oil loss occurred at a well in the Bay Ste. Elaine Camp (Menzel, 1948). This loss was about 25 barrels of crude oil which floated into the shore and into a small cove. At low tide this oil was deposited on the mud and also on a small intertidal oyster reef of coon oysters. There most of the oil adhered to mud, shells, and oysters. It shortly assumed the weathered brown appearance. This oil loss occurred on January 3, 1948. A month later the oysters were still covered with obvious brown sticky oil. Oily taste in these oysters remained until February 28, but by March 7 it was no longer detectable, and did not return. On January 19 the boxes amounted to 8.5 percent and on May 9, four months later, the count showed 5.8 percent of boxes.

Exact mortality data were secured on this occasion. One thousand oysters were collected from the oily mud on January 15 and placed in Sea Rac trays. These were divided into two groups, from one of which the oily mud was removed by scrubbing. These two groups of 500 oysters each were removed to Bayou Bas Bleu and observed until March 16. They both remained oily tasting until February 24. Mortalities occurred as shown in Table 53. The two groups are reported as one since the total mortality differed less than 1.0 percent in the two groups.

TABLE 53

Mortality of oysters covered with crude oil by oil loss of January 3, 1948, at Bay Ste. Elaine Oil Field, and removed to Bayou Bas Bleu on January 19, 1948

Date of examination	Mortality percent
January 20	0.10
January 25	0.45
January 30	0.45
February 4	0.30
February 9	0.50
February 14	0.50
February 19	0.00
February 24	0.20
February 29	0.20
March 16	0.10
Total mortality	2.10

An oil loss estimated at 800 barrels took place on November 5, 1951. This one was from Well 3–8 of The Texas Company's Bay Ste. Elaine Field. This well is one of a large number in a system of canals, bayous, and small bays in the marshes above Bay Coon Road. The oil lost and its effects were studied closely until all oil had disappeared. After the loss the oil spread rapidly into the system of canals, bayous and bays to the north and west of the well under impetus of a strong flood tide and an equally strong southeast wind.

On November 7, a strong northwest wind set in, at the beginning of an equatorial tide period. In the next several days tide level declined and varied, between high and low, only a maximum of 0.6 foot and on November 8 only 0.1 foot. During this period much of the oil disappeared; it is presumed to have been carried out into Terrebonne Bay

proper via Bayou Little Caillou. The oil was not observed to go out, since much of the ebb tide period was at night. Oil was not observed in Terrebonne Bay or any of the bays intervening to the Gulf, although efforts were made to locate oil to the south of the spilled area. But the very great decrease in the canals and bayous near where the loss occurred apparently meant that flushing into the Gulf occurred during the period of high north wind.

Some of the oil unquestionably evaporated from the water surface, and some was pocketed in small ponds and embayments in the marshes. It is estimated that losses to the Gulf by flushing, evaporation, and pocketing in small marsh ponds all together removed at least 90 percent of the original 800 barrels from observable parts of the canals and bayous. The remainder of the oil adhered to the mud in the intertidal zone and attached to marsh plants. The canals and bayous in the affected zone were estimated to have at least 30 miles of shoreline. The seventy-five to one hundred barrels of oil remaining in the affected zone was distributed along these shorelines, partially in the marsh and partially in the intertidal zone. One week after the loss this oil was still visible in patches along the shore with some thick layers in protected places. A more or less continuous oil slick covered most of the canals and portions of interconnecting bayous and small bays as shown by the map of November 11, 1951. Oil on that date apparently reached its greatest distribution, covering a linear distance of slightly over two miles east and west and about one and one-half miles north and south. It was observed on that date that light slicks were emerging from the shallow marsh ponds and the marsh itself, with the ebbing tide. From that date the surface slicks rapidly decreased, and it was noted that with a flood tide the canals largely cleared of oil, indicating that it was carried into the marshes and marsh ponds.

On November 16 there was a strong north wind, which continued through the 17th and shifted to the west on the 18th. By the latter date there were only small visible patches of oil on the water, in protected places. By this date weathering was largely complete and the oil, when found, was in the form of gummy brown encrustations. The "oil line" on the bordering marsh plants was still clearly visible over most of the area. There were occasional recurrences of oil slicks on the water when the wind stirred the mud in the intertidal zones.

Disappearance of the visible oil did not mean that the oil was gone from the area. Portions of it sticking to the bottom along the shores could easily be found by stirring the mud in the intertidal zones, and by shaking marsh plants (*Spartina* mostly). A series of stations was set up by choosing those areas where large amounts of oil were observed to be pocketed along the margins. The history of one of those stations is chosen for illustrative purposes because (1) the oil lasted longest there, and (2) there were coon oysters along the shore.

This station, Number 2, was located in the margin of a shallow natural bayou about one quarter mile from the site of the original loss. Visible oil was gone from this station by November 18. However, oil could be stirred from the bottom as late as January 25, 1952. On that date, vigorous stirring produced only a slight iridescence.

The original oil at Station 2 was a thick layer about one-half inch thick. This accumulated in the first period of the oil loss. The shore line at Station 2 was so angled that the oil was piled on shore at a time when evaporation had begun to reduce it to a brownish tar-like sheet. It adhered firmly to the bottom in the intertidal zone and extended into the marsh for a distance of at least ten feet, discoloring the basal part of the *Spar-*

tina stand. At no time was it possible to stir oil from the bottom below low tide level at this station. This was true also of all other stations. This does not mean that no oil at all reached the bottom below low tide. It does mean that the amount was so small as to be insignificant.

The original thick layer of oil thinned out very rapidly. How much was removed and dispersed by water current and how much was oxidized by bacterial action is not known. By November 18 the oil layer as such had disappeared, but heavy slicks could be stirred from the bottom at that date. Thereafter the amount rapidly decreased; the last date on which any iridescence could be produced at Station 2 was January 25, 1952. That was just a little less than three months from the time of the original oil loss.

At Station 2 and located in the middle of the heaviest oil was a small oyster reef, not more than a dozen feet in diameter. When oil was at its thickest, those oysters lying flat on the bottom were completely covered. About a dozen new boxes were recovered at this time from this small reef. They were believed to have been smothered by the oil, and thus bear the distinction of being the only oysters ever observed to be killed by accidental oil loss. Those oysters standing upright were unharmed. Repeated checks of this small reef until the end of observations in late January, 1952, showed rapid growth and no additional mortality. Oil could be observed sticking to the oysters for about two months after the oil loss.

Oily-tasting oysters were found at several stations. Those at Station 2 retained an oily taste to January 25, 1952, which was 81 days after the oil was spilled. At all other stations the period of retention of oily taste was shorter.

At only one point were subtidal oysters found in the area where oil was observed. This was on an oyster lease in Ray Bayou approximately ¾ mile north of the site of the oil loss. In this area oysters do not grow naturally below low tide, and the death rate of subtidal planted oysters is about 60 to 80 percent per year if they remain through a warm season. The oysters found were remnants of a planting which probably had been harvested. Enough live oysters were found to permit sampling for oily taste. Oil was observed over this bed and the *Spartina* was marked with a heavy oil line. However, no oily-tasting oysters were ever found here, even when the oil was over the bed, as it was when the first samples were taken (November 11).

A close check of the fauna and flora was made during the three months of the study. At Station 2, where smothered oysters were found, a number of dead mussels, *Brachidontes recurvus*, with the meats still intact were also found. So far as could be determined no other species of animal or plant was affected. *Polydora, Cliona, Membranipora, Thais, Eurypanopeus, Callinectes, Balanus*, gobies, and a few toad fish were found associated with the oysters. Algal growths were unaffected. In the marsh, there were thousands of *Littorina, Uca*, and a few *Sesarma*, which did not move out of the oily area. Goby eggs were particularly numerous attached to the oyster shells, especially in boxes.

Studies on Glycogen in Oysters Treated with Oil and Bleedwater

In several experiments using oil or bleedwater, glycogen analyses were made on oysters at the end of the study, comparing controls and experimental oysters. The results of studies in the Grand Isle Laboratory and the Barataria Bay area are presented in Table 54, and the results of studies in Bay Ste. Elaine and Lake Barre oil fields are summarized in Table 55. For Table 54, the references are: Mackin, 1948a, page 18;

TABLE 54

Summary of studies on the effect of oil or bleedwater on glycogen content of oysters in the Barataria Bay area

Oil or bleedwater application	Glycogen, percent of wet weight		Remarks
	Experiment	Control	
Oil sprayed on oysters in field for 6 months	3.78	6.10	Only 8 oysters analyzed
Oil sprayed on oysters in field for 6 months	4.19	5.36	About 60 oysters in each sample
Emulsified oil applied in laboratory for 6 months	0.174	0.292	Inadequate number of survivors for dependable check
Heavy applications of emulsified oil over a period of 8 days to 2 weeks; observed for 6 or 7 months in field	8.62	7.89	60 oysters in each sample
Bleedwater in concentrations of 1% and 2.5% applied in aquaria for 6 months	1.47	1.40	Aquaria treated with bleedwater developed heavy blooms of diatoms and other algal forms
	1.62	0.815	
Means	3.252	3.643	.....

TABLE 55

Summary of field studies on the effect of bleedwater on glycogen content of oysters in oil field areas

Study No.	Period studied	Distance from BW outlets	Glycogen, percent of wet weight	
			At start	At end
B. Ste. E. No. 1	Dec. 1947–Jan. 1949	200 ft	2.29	5.04*
		1 mile	3.53	3.88*
B. Ste. E. No. 2	Aug. 1947–Aug. 1948	Between outlets	0.93	1.66
		2160 ft	0.93	1.20
B. Ste. E. No. 3	Sept.1948–Apr. 1949	Between outlets	.....	5.59
		2160 ft	.....	6.64
L. Barre No. 1	Jan. 1948–Aug. 1948	80 ft, top trays	3.04	1.32
		80 ft, bottom trays	1.85	1.56
		3200 ft, top trays	3.84	1.08
		3200 ft, bottom trays	3.73	1.22
L. Barre No. 2	Oct. 1948–May 1949	25 ft north	.....	0.557
		50 ft south	.....	0.568
		100 ft south	.....	1.540
		150 ft south	.....	1.740
		3200 ft south	.....	2.370

* These commercial bottom plantings were sampled monthly; glycogen percent fluctuated from 1.29 to 6.70 in the experimental plot and from 1.02 to 5.02 in the control plot. This may be compared with the range from 1.71 to 6.11 in Bayou Bas Bleu during the same period.

1948b, page 3; 1948e, page 5; 1949, page 4; 1950a, page 3. For Table 55, the references are: Menzel and Hopkins, 1951, pages 62, 95; 1952, page 124; 1953, pages 59–60 and 144, 149.

STUDIES ON THE EFFECT OF NATURAL GAS FROM DEEP WELLS ON MORTALITY, GROWTH, AND GLYCOGEN CONTENT OF OYSTERS

As natural gas leaking from pipe lines had been mentioned by some oystermen as a possible cause of oyster mortality, three field tests on the effects of deep well gas were conducted at Bay Ste. Elaine oil field by Menzel (1949). Waste gas here was discharged

under water at the rate of five cubic feet per minute. Oysters in wire baskets were placed at the gas outlet and at various distances. The mortality, growth, and change in glycogen content of experimental and control oysters are shown in Table 56. For details see the original report (Menzel, 1949).

TABLE 56

Effect of natural gas on mortality, growth, and glycogen content of oysters

Study no.	Test period		Mortality percent	Percent increase in shell length	Glycogen, percent of wet weight	
					Start	End
1	Aug. 1947–	Exper.*	9.6	80.8	.....	.....
	Jan. 1948	Control†	7.0	37.3	.....	.....
2	Apr. 1948–	Exper.‡	41.7	13.9	.....	.....
	Sept. 1948	Control†	42.5	20.5	.....	.....
		Exper.§				
3	Mar. 1949–	Group I	8.0	—1.12	5.50	2.75
	May 1949	Group II	22.7	3.09	2.97	2.87
		Control‖				
		Group I	8.0	0.25	5.50	4.20
		Group II	16.0	6.51	2.97	3.39

* Averages of baskets under gas exhaust, at four locations 10 feet away, and at four locations 25 feet away.
† Control baskets were 3500 feet north of gas outlet.
‡ Averages of one basket under gas exhaust and four 10 feet away.
§ Experimental oysters were in a wooden box with hardware cloth top and bottom, enclosing the gas outlet so that gas bubbled up through the oysters. The two groups were of different sizes and origins.
‖ Control oysters were in an identical box without gas 36 feet away.

EXPERIMENTS WITH COMPONENTS OF DRILLING MUD

In rotary drilling, a heavy fluid mixture known as drilling mud is pumped down through the hollow drill stem to cool the bit, coat the walls of the well, and prevent gas blow-outs; it returns to the surface through the space between the drill stem and the walls of the well, bringing with it the rock cuttings from the bit. After cuttings are screened out for geological study, the used drilling mud from a barge-mounted drilling rig may be dumped into a mud barge for disposal or to be processed for reuse, or it may be dumped overboard. Drilling mud is therefore among the products of oil operations that have been suspected of killing oysters.

In a study of the effect of barium sulphate, the principal chemical constituent of drilling mud, one sack of Magcobar was used to bottom the experimental aquarium (60 by 95 cm) to a depth of about 4 to 5 cm. Water covered the Magcobar to a depth of 10 cm, and experimental oysters were placed on the drilling mud. A mud-sand mixture bottomed the control tank. The water flow was 500 cm per minute in both tanks (Table 57).

TABLE 57

Results of drilling mud experiment, testing the effect of barium sulphate on oysters, December 10, 1949 to April 10, 1950
Reference: Mackin, 1950d

	No. of oysters	Number dead	Percent dead
Experiment	30	2	6.7
Control	30	3	10.0

Quebracho, powdered bark from the "axe breaker" tree of South America, is used in the leather tanning industry for its high tannin content. It is also an important constituent of many drilling mud mixtures. Six laboratory experiments to test the effect of quebracho on oyster survival were carried out at Grand Isle. A stock solution of 100 grams of quebracho (dry powder) in 20 liters of sea water was prepared. This gave a dark wine-red mixture, part solution and part suspension of fine particles. The stock solution was dripped into experimental aquaria in controlled amounts and the sea water flow was maintained at 2000 cc per minute in all aquaria. Experiments were run in two sets of three, each set with one control (Table 58).

TABLE 58

Tests of the effect of quebracho on mortality of oysters
Reference: Mackin, 1951b

Concentration of Quebracho	No. oysters used	Number dead	Percent dead
Results of first set of three experiments, August 6, 1950 to September 21, 1950			
1/10 stock	15	9	60.0
1/20 stock	15	8	53.3
1/40 stock	14	6	42.9
Control	15	8	53.3
Results of second set of three experiments, October 9, 1950 to June 9, 1951			
1/10 stock	15	5	33.3
1/5 stock	15	5	33.3
2/3 stock	15	10	66.7
Control	15	5	33.3

STUDIES ON SEISMOGRAPHING AND ITS EFFECT ON OYSTERS

The first studies of the effect of seismographic exploration on oysters were described in the report of the Louisiana Department of Wildlife and Fisheries for 1938–1939 (McConnell, 1949). The experiments were carried out by L. D. Kavanaugh of the Division of Water Bottoms. Details were not given, except that the shots were on the bottom, at distances of 175 to 750 feet from experimental oysters, and shots of 30 to 70 pounds of "high velocity" gelatine were used. These studies were not conclusive, and mortality data were not given.

McConnell (1944) stated that the seismic division had investigated the effects of seismic exploration further, but no report has been published. Gowanloch and McDougall (1944) found that 800 pound shots on the bottom failed to harm oysters at 50 feet. The same authors (1945) again set up experiments to test the effects of heavy charges (up to 400 pounds). These charges were sunk 25 feet below the surface of an oyster bed. No mortality resulted in the six weeks that experimental oysters were kept under observation.

Sieling (1954) quoted an anonymous report of the Chesapeake Biological Laboratory which stated that 2 percent of oysters exposed in bags on the bottom within 100 feet of a 30 pound suspended charge, and within 200 feet of a 300 pound charge were killed immediately and 5 percent were fatally injured.

Kemp (1954) found that 40 pound shots placed 20 feet below the bottom killed oysters as far out as 200 feet. The data from the experiments reported by Kemp appear to be contradictory: in the study which showed that oysters were affected at 200 feet,

fish, shrimp, and blue crabs were unaffected at 10 feet; in another study where oysters were said to be affected at 50 feet, fish, shrimp, and blue crabs were not affected at less than 5 feet (at the shot point). Kemp's results seemed to directly contradict Gowanloch's.

Studies by the Research Foundation

Experiments by the Research Foundation were designed to test immediate and/or delayed effects of seismographic exploration on oysters. Effect of shock, effect of gases liberated from the bottom, and effect of explosives in causing oysters to sink in soft bottom, were all investigated by Sieling (1951, 1954). He set up two studies, one in Bay Baptiste, a northeast extension of Barataria Bay, and the other in Hackberry Bay, just to the northwest of Barataria Bay. Oysters were held in trays, which were placed at measured distances of 20, 60, 130, and 250 feet from a shot point in a staggered line. This shot point was flanked in four directions by additional shot points 1000 feet from the central one. Controls were set up 750 feet from the nearest shot point. Bottom plantings were made at 40 feet from each central shot point, in staked areas 18 feet square.

Distant controls were placed at Bay Chene Fleur and in mid-lower Barataria Bay. The Chene Fleur station had a lower salinity than the experimental areas and the Barataria Bay stations were in higher salinity waters.

Explosive charges of 50 pounds at 70 feet deep, and 20 pounds at 30 feet deep, were discharged at each shot point. The five stations of a shot point group were fired successively, as fast as the field crew could make the necessary connections. Trays at the Bay Baptiste experimental and control stations were placed on the bottom, while at Hackberry Bay the tray oysters were held on racks.

Chemical measurements were made immediately before the experimental shots, immediately after the shots, and at intervals for several months. Oysters at each station were examined immediately before and immediately after the shots to determine whether shock effects were present. One day later, part of the oysters of each control station were moved to the experimental area (5 trays, one at each measured distance from the shot point). Similarly, trays from the experimental stations were moved to the control stations. These shifts were designed to detect delayed effects, as distinguished from shocks effects, if any were present. Checks at experimental and control stations were made thereafter at approximately two-week intervals for eight months, except for some trays which were stolen from Hackberry Bay station.

Results of these studies are presented below in Tables 59 and 60 from Sieling (1954).

Table 59

Results of experiment testing the effect of seismographic exploration on oysters in Hackberry Bay

Distance from central shot point	No. oysters started*	No. oysters surviving 4 months†	Percent survival 4 months	No. oysters surviving 8 months‡	Percent survival 8 months
Rack #1, 20 feet					
Permanent Trays	345	289	83.7	.....	.....
From Chene Fleur	172	157	91.2	.....	.....
From Station 51	155	92	59.3	.....	.....
Rack #2, 60 feet					
Permanent Trays	348	302	86.7	.....	.....
From Chene Fleur	172	152	88.3	.....	.....
From Station 51	166	100	60.2	.....	.....
Rack #3, 130 feet					
Permanent Trays	336	287	85.3	.....	.....
From Chene Fleur	170	147	86.5	.....	.....
From Station 51	156	73	46.8	.....	.....

Rack #4, 250 feet					
Permanent Trays	333	281	84.4	.....	.....
From Chene Fleur	170	156	91.7	.....	.....
From Station 51	162	96	59.2	.....	.....
Controls in Hackberry Bay					
Permanent Trays	338	257	76.0	.....	.....
From Chene Fleur	169	133	78.6	.....	.....
From Station 51	163	86	52.8	.....	.....
Controls at Chene Fleur					
Permanent Trays	864	789	91.3	761	88.1
From Rack 1	172	150	87.2	136	79.1
From Rack 2	171	148	86.8	141	82.5
From Rack 3	168	152	90.4	144	85.7
From Rack 4	173	151	87.3	143	82.6
From Control Rack	171	146	85.4	140	81.9
Controls at Station 51					
Permanent Trays	825	625	75.8	567	68.7
From Rack 1	164	139	84.7	123	75.0
From Rack 2	169	143	84.6	132	78.1
From Rack 3	169	137	81.0	121	71.6
From Rack 4	169	132	78.1	116	68.7
From Control Rack	166	142	85.5	126	75.9

* Permanent trays at Station 51 started 8–20–49, trays at Chene Fleur started 8–19–49, all other trays started 8–23–49.
† Dates taken as end of 4 months period were 1–5–50 for controls at Station 51 and 1–9–50 for all other trays.
‡ Dates for end of 8 months period were 4–22–50 for Station 51 and 4–23–50 for Chene Fleur.

TABLE 60

Results of experiment testing the effect of seismographic exploration on oysters in Bay Baptiste

Distance from central shot point	No. oysters started*	No. oysters surviving 7½ months†	Percent survival 7½ months
Rack #1, 20 feet			
Permanent Trays	334	253	75.7
From Chene Fleur	168	134	79.8
From Station 51	143	105	73.4
Rack #2, 60 feet			
Permanent Trays	326	256	78.5
From Chene Fleur	164	131	79.9
From Station 51	127	111	87.4
Rack #3, 130 feet			
Permanent Trays	324	255	78.7
From Chene Fleur	166	127	76.5
From Station 51	151	108	71.5
Rack #4, 250 feet			
Permanent Trays	329	242	73.5
From Chene Fleur	164	129	78.6
From Station 51	148	111	75.0
Control Bay Baptiste			
Permanent Trays	341	264	77.4
From Chene Fleur	170	133	78.2
From Station 51	150	118	78.6
Controls at Chene Fleur			
Permanent Trays	834	736	88.2
From Rack 1	159	139	87.4
From Rack 2	166	138	83.1
From Rack 3	128	97	75.7
From Rack 4	165	143	86.7
From Control Rack	171	139	81.3
Controls at Station 51			
Permanent Trays	820	596	72.7
From Rack 1	164	121	73.7
From Rack 2	167	112	67.0
From Rack 3	167	104	62.2
From Rack 4	166	120	72.2
From Control Rack	170	109	64.1

* Permanent trays at Chene Fleur started 8–30–49, all other trays 8–31–49.
† 7½ months ended 4–22–50 for Station 51, 4–23–50 for Chene Fleur, and 4–21–50 for Bay Baptiste.

These experiments showed no effect of shock, no delayed mortalities, and no change in the bottom caused by the explosions. The numbers of oysters used were great enough that there is no question as to the statistical reliability of the studies. Kemp's studies cited above were definitely faulty in that respect. Sieling found no immediate gaping of oysters checked immediately after the shots were fired. The period of observation was longer than in any previous test of the effect of explosions. However, in Gowanloch's study the oysters removed to a distant station after the shots were observed long enough for practical purposes.

Summary and Conclusions

1. The history of the oyster industry shows that Louisiana oysters have always been subject to high rates of mortality, and that periods of disastrously high mortality have been frequent as far back as the records go.

2. Louisiana oyster production statistics indicate that the period 1940 to 1947, when catastrophic mortality caused by oil operations was alleged in law suits, was in fact the most productive eight-year period in the history of the Louisiana oyster industry. The official figures show a mean annual production of more than 850,000 barrels of oysters during this period.

3. The same figures show that oyster production declined in the state as a whole during the period 1940 to 1947, so that the last four years (1944 to 1947) had an average annual production of only about 635,000 barrels. A sharp drop in production in St. Bernard Parish, according to the official Louisiana statistics, was responsible for the greater part of this decline. St. Bernard Parish is far outside of the area where damage from oil operations was claimed.

4. The study of Louisiana oyster production statistics and oyster history showed that disastrously high mortality of oysters had occurred at times both before and after oil production began in the oyster-growing area, and that since oil production started there had been oyster mortalities in places far distant from oil operations as well as in and near oil fields.

5. Numerous accounts in the literature, including official reports of Louisiana state agencies concerned with management of the oyster industry, have linked high mortalities in the past with high salinity of the water over the beds.

6. Field studies of Texas A&M Research Foundation biologists, beginning in 1947, confirmed reports that mortality rates were high on many Louisiana oyster beds, and that there was a seasonal cycle in mortality correlated with temperature. Except during abnormally warm periods there was little mortality in winter, but oysters began to die in spring and continued to die steadily all summer and into autumn until stopped by cool weather. The regular and predictable nature of this mortality indicated that it was not abnormal. The general picture was, rather, that a high rate of mortality associated with summer temperatures was normal in much of the Louisiana oyster-growing territory.

7. Field studies of Foundation biologists also showed that within the region where damage from oil operations was claimed (in general, Placquemines, Jefferson, Lafourche, and Terrebonne Parishes) there were areas where oyster mortality was consistently low as well as areas of high mortality. No correlation was found between rates of mortality of oysters and their proximity to oil fields. Indeed, in the Barataria Bay area where most damage to oyster production was claimed, the highest mortalities were

found at the stations farthest from centers of oil and bleedwater production. On the other hand, high mortality was found to be correlated with high salinity of the water.

8. Extensive studies by Texas A&M Research Foundation chemists have shown that measurable amounts of substances which are extractable with carbon tetrachloride, and are unsaponifiable, are present in natural muds of water bottoms far removed from any source of oil pollution as well as near oil fields. Unsaponifiable carbon tetrachloride extractives have been called "hydrocarbons" by some chemists without further attempts at identification. However, Foundation chemists found that the quantity of so-called "hydrocarbons" was correlated with amount of plant debris in samples, and that such substances could be extracted from aquatic plants.

9. The quantity of so-called "hydrocarbons" in mud samples taken from bottoms, including oyster beds claimed to be damaged by pollution, in high mortality areas was no greater than in muds from low mortality areas.

10. Studies of mud samples taken in oil fields at and near bleedwater outlets, in some cases showed quantities of unsaponifiable carbon tetrachloride extractives far greater than any found elsewhere, and these samples were also shown by Horvitz to have quantities of substances identified as "pentane and heavier hydrocarbons" greater than any found at points distant from oil production centers. However, these quantities diminished rapidly with distance away from points of effluent discharge. In no case was oil from a bleedwater outlet traced to an oyster bed, although intervening bottoms were sampled diligently.

11. Analyses of many water samples also showed that substances identifiable by uncritical routine analysis as "hydrocarbons" are found in minute quantities in Louisiana bay waters, and that quantities of such substances are no higher in areas of high oyster mortality than in low mortality areas. Much larger quantities are found in bleedwater effluents, but in natural waters into which the bleedwater discharges the amounts are negligible, rapidly decreasing with distance away from the point of discharge and soon dropping to the barely detectable level found in all bay waters.

12. Calculations based on the actual production of bleedwater in Louisiana oil fields, and the volumes of water in bays, have shown that oil from this source could not possibly reach a significant level of concentration in oyster-growing bay waters, or in bottoms of these water bodies, even if none were destroyed.

13. Bacteriological studies have shown that crude oil and fractions of crude oil are rapidly oxidized and destroyed by bacteria which live in Louisiana bay muds.

14. Studies of a few cases of accidental spillage of crude oil in oyster-producing waters have shown that such oil losses may cause an oily taste in oysters in the vicinity, but the oysters rid themselves of the oily taste in a few days or weeks, depending on the degree of contamination and other factors. With the exception of one case where a heavy oil deposit apparently smothered a few intertidal oysters, no mortality of oysters resulted from the oil losses studied.

15. The only studies by any previous investigator that indicated oysters could be killed by small amounts of crude oil or oil extracts were those by Gowanloch in 1933. Biologists of the Texas A&M Research Foundation carefully repeated Gowanloch's laboratory experiments and failed to confirm his results. The survival of oysters was not affected by oil in exact duplicates of Gowanloch's experiments.

16. Other laboratory studies by Foundation personnel on the effect of bleedwater, crude oil, and extracts (so-called "soluble fraction") of oil on survival of oysters, using concentrations of pollutants greater than could occur on oyster beds, have given uniformly negative results.

17. Laboratory studies by Foundation personnel and consultants on the physiological effects of crude oil, "soluble fractions" of crude oil, and bleedwater have shown that a detectable effect on activities of oysters is not produced by any concentration of oil or bleedwater that can occur in oyster-producing waters (bays or bayous) of Louisiana. Physiological effects (slowing of water pumping and filtering activity) were caused by amounts far in excess of any concentration possible in the waters over an oyster bed. These effects did not result in harm to the oysters, and were completely reversible.

18. Field experiments in Barataria Bay showed that spraying oysters repeatedly with crude oil and keeping oysters under a surface layer of crude oil did not affect survival or growth.

19. At Lake Barre Oil Field, where the amount of bleedwater discharged directly into an oyster-producing water body was greater than at any other place in Louisiana, field experiments showed that oysters were killed by bleedwater. However, this lethal effect extended only to a distance of 50 to 75 feet from the point of discharge. Shell growth and accumulation of glycogen in oysters were adversely affected to a distance of approximately 150 feet from the bleedwater outlets, but there was no harmful effect at greater distances.

20. Other experiments, including field experiments on setting, growth, survival, and fattening of oysters at various distances from bleedwater and waste gas outlets in various oil fields, failed to show any harmful effect of oil, bleedwater, or natural gas.

21. Laboratory experiments showed that barium sulphate and quebracho, principal constituents of drilling muds, have no lethal effects on oysters in concentrations that could be maintained in water over an oyster bed. Barium sulphate is insoluble and inert, and has no toxic properties. Quebracho is toxic at concentrations higher than those possible in natural waters.

22. Field experiments showed that high explosives discharged in holes drilled in the bottom, in the manner prescribed by Louisiana law for seismographic exploration in enclosed waters, did not affect oysters in any way.

23. We conclude from the history of Louisiana oyster production, and from Foundation studies in the field and laboratory, that oil operations could not have been responsible for widespread mortalities of oysters as alleged. Those widespread mortalities which actually occurred during the period of our studies resulted from moderately but persistently high daily mortality rates which were associated with high temperatures and high salinities, and were preventable by decrease in either temperature or salinity, entirely independent of oil operations.

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Oyster Disease Caused by *Dermocystidium marinum* and Other Microorganisms in Louisiana[1]

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[1] Oceanography and Meteorology Series from the Department of Oceanography and Meteorology, the A. and M. College of Texas. A report from Project 23E, Texas A. and M. Research Foundation, sponsored by Texaco Incorporated, Humble Oil and Refining Company, The California Company, Gulf Refining Company, Phillips Petroleum Company, and Shell Oil Company.

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Abstract

When the causes of extensive summer mortality among oysters (*Crassostrea virginica* Gmelin) in Louisiana could not be traced to oil operations or other environmental causes, a study was made of the diseases, especially the fungus, *Dermocystidium marinum* Mackin, Owen, and Collier, its general biology, systematic position, distribution, seasonal patterns, eipizootiology, and significance in oyster populations and the oyster industry. This is a summary of experiments, field studies, and histological work in Texas A & M Research Foundation Projects 9 and 23 providing an ecological view of the environmental and parasite-host relations of a disease in a marine shellfish population.

Dermocystidium marinum Mackin, Owen, and Collier, a fungus parasite of oysters develops by radial cleavage from small unicellular thalli to multinucleate plasmodia or sporangia, in intracellular position in oyster host cells, or free in blood. Occasionally cells of the parasite appear to bud. The parasite may repeat the reproductive cycle or develop into pre-hypnospores, cells with a large vacuole containing volutin, presumed to develop into resistant winter hypnospores or sporangia when liberated into sea water on death of the host.

Crude cultures of *D. marinum* with *Fusarium* were accomplished in which rapid reproduction of the cells took place for up to about 30 days. Sea water was used for a medium. No subcultures could be made from these cultures but infection of oysters was accomplished from them. All of these crude cultures ended by production of aplanospores from sporangia or sori of sporangia. It is believed that the normal life cycle of *D. marinum* may involve a short reproductive cycle in sea water, which ends by production of non-motile spores in sporangia. Based upon structure, cytochemical characteristics and developmental cycle, *D. marinum* is placed provisionally in the family Rhinosporidiaceae Ciferri, and it is suggested that the genus *Dermocystidium* and a number of other genera of low parasitic fungi form a group coordinate with the Olpidiaceae and other chytrids. These apparently related genera agree in the production of plasmodia (or sporangia ?) by repeated nuclear divisions and cleavage.

Dermocystidium marinum is discontinuously distributed in the estuaries of the Gulf of Mexico and Atlantic coast from the vicinity of Tampico, Mexico to Delaware Bay. In Louisiana the fungus is reported from more than one hundred bays and bayous, in salinities from about 9 ppt up to oceanic level. The distribution in Louisiana is about coincident with that of the oyster host.

Experimental studies show that infection by *D. marinum* is usually lethal to the host oyster in high temperature periods. It was demonstrated also that physiological damage also results from infection. Weakness of oysters in advanced states of infection was demonstrated and it was also shown that loss of weight, castration, and failure to grow normally may be a result of infection. Histopathological studies showed extensive lysis of tissues, destruction of Leidig cell connective tissue, reduction of glycogen, and abnormal production of ceroid. Defensive reaction of the host consists of phagocytation and removal of *D. marinum* cells by amoebocytes, which may leave the host by any epithelial surface.

Epizootics under natural conditions, in oysters held in trays, and in aquaria were studied. Temperature directly controls the development of epizootics which reach their apex in the summer or early fall. In subtropic areas *D. marinum* infections may carry through the winter; but in the Chesapeake and Delaware Bay populations of oysters lose all apparent infection. Low salinity areas (under 9 ppt) usually have few or no infections. Above S 10 ppt, incidence of infection of populations increases. It is believed that flushing of estuaries may be more effective in decreasing infection rates than is low salinity *per se*.

An examination of several control methods for *D. marinum* indicates that it is possible, by utilizing environmental factors, seasonal variation in death rates and apparent immunity in young oysters, to successfully cultivate oysters in areas with high infection rates. Chemical control and use of immune races have as yet not proved feasible, but may be of value later.

In addition to disease caused by *D. marinum* several other diseases and parasites are described. These are "mycelial disease," "amber disease" caused by an amoeboid organism, yellow pustule disease, watery cysts, a bacterium parasitizing the gastric shield, *Nematopsis*, and a ciliate parasite of the digestive tract. The effect of these diseases and parasites on the host oyster is still unknown.

Introduction

As shown in the preceding paper in this volume (Mackin and Hopkins, 1962), mortality of oysters in Louisiana was often very high, ranging from 30 percent to 100 percent per year. Data were also presented which indicated that pollution was not responsible, and that conchs, crabs, and other predators, while locally abundant and

capable of producing high mortality rates within limited areas, did not cause the widespread heavy losses of mature oysters observed in many areas of Louisiana. The greater part of the excessive mortalities occurred in waters relatively free of predators.

It seemed certain, therefore, that there was a cause of mortality in Louisiana oysters which involved neither pollution nor predators. Because of the report of Prytherch (1938a, 1940) that *Nematopsis ostrearum*, a gregarine sporozoan, was highly destructive to oysters, this parasite was thoroughly studied by Dr. Victor Sprague (1949, 1950), and Sprague and Orr (1953), with the result that these authors were unable to confirm Prytherch's claims (see below, page 204). Attention was soon turned to a search in oysters for new disease-producing organisms, since it was obvious, in the spring and summer of 1948, that oysters were dying in large numbers. These studies led to the discovery of *Dermocystidium marinum* (Mackin, Owen, and Collier, 1950). From 1948 to the present, further studies have been designed to throw light on the morphology, life cycle, pathogenicity and epizootiology of this fungus parasite. It is the aim of this paper to summarize the known data relating to *Dermocystidium marinum*, and the disease of oysters caused by it.

Several other diseases and parasites of oysters were also encountered in Louisiana Bays. Like *Nematopsis*, some of these cause no mortality of oysters, but others may be lethal under certain conditions. Data on these additional parasites and diseases are also reported.

Biology of *Dermocystidium marinum*

History of Researches on *Dermocystidium marinum*

In Project Nine studies at Grand Isle, Louisiana, oysters which were obviously diseased were observed in numbers in the spring of 1948. At that time and during the following summer numerous aquarium studies were carried on, and as the weather warmed after an exceptionally cold winter, deaths occurred in aquaria in considerable numbers. It became a part of the routine to make smears of blood and teased-tissues of the gapers. Sick-appearing oysters were found occasionally in the field. A great variety of protozoa, bacteria, and fungi were observed in the smears made from oysters with abscesses and shrunken meats. Of all the organisms associated with dying oysters, one was regularly observed in most gaping oysters. Its intra-cellular position indicated that it was present prior to the time the oyster gaped, and was probably a true parasite. This organism was the one later named *Dermocystidium marinum*.

At about the same time Mr. Albert Collier and his associates made parallel observations at Pensacola, Florida. Mr. Collier in these early studies believed that the organism might be a colorless *Chlorella*, while we thought it might be an odd yeast, both hypotheses being based on the large eccentric vacuole of the hypnospore stage.

Little more was accomplished in the summer of 1948. During the fall, several studies by Mr. J. Prokop were attempted to determine if bacteria were involved in oyster disease, with negative or inconclusive results.

Dr. Malcolm Owen (1955), working for the Division of Oyster Bottoms of Louisiana, had made observations of epidemiological nature which led him to hypothesize a new disease of oysters. Dr. Owen in early 1949 was shown prepared slides of *D. marinum* and brought up to date on Research Foundation studies.

The earliest report on the disease was by Mackin and Wray (1949). Here the causative organism was described as a "yeast-like" fungus, and some meagre details of histopathology and symptomology were given. Mackin, Owen and Collier (1950) described the oyster parasite as *Dermocystidium marinum*, presented some details of cytology, discussed the genus *Dermocystidium* Perez 1908, and called attention to the association of the fungus and mortality of oysters in Louisiana. Mackin (1951a) stated that the disease caused by *D. marinum* was lethal to oysters, and was a major factor in destruction of oysters in Louisiana. The results of studies of histopathologic effects of the fungus on its host were reviewed by Mackin (1951b).

In the early part of the studies on *D. marinum*, the taxonomic relations were not known. During the latter part of 1948 and early 1949, Dr. S. H. Hopkins studied literature on various obscure protistans. *D. marinum* was placed by Mackin, Owen and Collier (1950) in the genus *Dermocystidium* when Hopkins located the papers by Perez (1907, 1908, 1913). This action failed to solve the problem of broad taxonomic placement, since *Dermocystidium* was itself a taxonomic orphan (a genus assigned to no major taxa). The organism was considered to be a highly aberrant form with both protozoal and fungal characteristics.

Mackin and Wray (1952) summarized the work of the Texas A&M Research Foundation on oyster disease, including results of all studies on structure, development, incidence, distribution, epizootiology, and history up to May 31, 1950, the termination date of Project Nine. Project Twenty-Three was organized in 1950 primarily to continue work on *D. marinum* and other disease-producing organisms.

Studies on *D. marinum* were also developed at Rice Institute at Houston, Texas by S. M. Ray. Ray's work at Rice Institute began in 1950 and eventually produced (1952a, 1952b, 1953) a rapid and reliable method for diagnosis of *D. marinum* infection in oysters. Ray's discovery gave great impetus to further studies. Ray also showed that *D. marinum* was a fungus, and collaborating with Mackin at the Grand Isle Laboratory, he improved methods for quick infection of the host oyster. These studies paved the way for accurate measurement of pathogenicity. The work is reported in several papers (Mackin, Ray and Boswell, 1953; Mackin and Boswell, 1953; Ray, 1954a, 1954b). Ray and Chandler (1955) reviewed the published data on *D. marinum*.

Ray (1954b) also demonstrated, using his diagnostic method, that organisms similar to *D. marinum* were present in two species of oysters other than *Crassostrea virginica*. These were *Ostrea frons* and *O. equestris*. *O. lurida* was reported by Ray (1954b) to have been artificially infected by J. R. Uzmann. Other marine animals in the Gulf area found by Ray to be naturally infected with *Dermocystidium*-like organisms were *Crepidula plana*, *Pecten irradians*, *Anomia simplex*, and *Martesia sp.* Ray and Chandler (1955) mentioned that certain nereid worms were sometimes found to be parasitized by a fungus appearing to be *D. marinum*. Mackin (1953d) found one nereid with an advanced infection with a *Dermocystidium*-like fungus.

Hewatt and Andrews began studies of oyster mortalities in the lower Chesapeake Bay and its estuaries in 1950 (Andrews, 1954, 1955b, 1956a, 1956b; Andrews and Hewatt, 1954, 1957; Hewatt and Andrews, 1954a, 1954b, 1956). Also Andrews (1955a) demonstrated that *Dermocystidium*-like fungi were found in twelve out of sixteen species of molluscan hosts examined in the Chesapeake area. This work suggests that *D. marinum* may be only one of a group of low marine fungi until recently unknown.

Ingle and Dawson (1953), reported the presence of *D. marinum* in oysters of

Apalachicola Bay, Florida. Dawson (1955) published details of a considerable survey of oysters of this bay and showed that the parasite was present at most of 19 stations studied, with a mean infection rate of 50 percent. Dunnington (1956) found *D. marinum* in various locations in the southern part of the Maryland Chesapeake, but no specific data have been published.

Dr. Harold Haskins and associates have studied recently the occurrence of *D. marinum* in Delaware Bay on the New Jersey side. As yet no data have been published but some data are contained in a mimeographed release of the New Jersey Oyster Research Laboratory at Bivalve, New Jersey (Anonymous, 1958).

Cytology, Life History and Development in Culture

Mackin, Owen and Collier (1950) presented a few details of the cytology of *D. marinum.* Mackin (1951b) published photomicrographs showing stages in reproduction and the vacuolate "spore" stages in oyster tissues. Mackin and Wray (1952) described the early cleavage progression in development, presented drawings illustrating these stages and photomicrographs of parasites in live smears and stained sections. Mackin and Boswell (1956) described a life cycle which involved a parasitic cycle of cleavage and development to the pre-hypnospore stage in the oyster host, and a hypothetical free development of the hypnospore in seawater to the sporangium with aplanospores.

In this section early development, cytology, and reproduction in seawater culture are discussed in some detail.

D. marinum in Live Smears from Oyster Tissue. In unstained preparations made from blood of heavily infected oysters, *D. marinum* is most often seen as a rounded mass contained within a leucocyte (Fig. 1: A,B). The protoplasm usually has a slightly bottle-green color, and nearly always there are several to numerous dark refractive granules contained in the cytoplasm. These granules are probably lipoidal in nature and are never seen in stained preparations (Fig. 1: E,F), since they are dissolved in ordinary chemicals used in the fixation, embedding, and staining techniques. Often several *D. marinum* cells or multinucleate stages may be contained in a single host cell (Fig. 1C). Multinucleate stages may be contained in host cells which also contain vacuolated uninucleate stages. Individual cells of multicellular stages at maturity represent the simplest form of the thallus. When one of these multicellular stages is gently pressed under the cover glass it can be separated into its daughter cells, each of which is wedge-shaped (Fig. 1D), with dense cytoplasm and one to several of the dark lipoidal granules.

Stages in the development of *D. marinum* as sketched from live smears (Fig. 2) sometimes resemble reproductive stages of yeasts very closely. These are not common but reproduction by budding is a normal process. Ordinarily a morula-like sphere of cells results from reproduction (Fig. 2: M,N). The number of cells resulting from cleavage may be as low as four (Fig. 2: O) or in excess of one hundred. When mature, the individual cells of an aggregation are clearly distinguishable (Fig. 2 N) radially arranged. It is believed that the structure is equivalent to a sporangium, but there may or may not be a common wall around the whole when it is contained in the host tissue.

The wedge shaped cells may round up prior to or after liberation from the

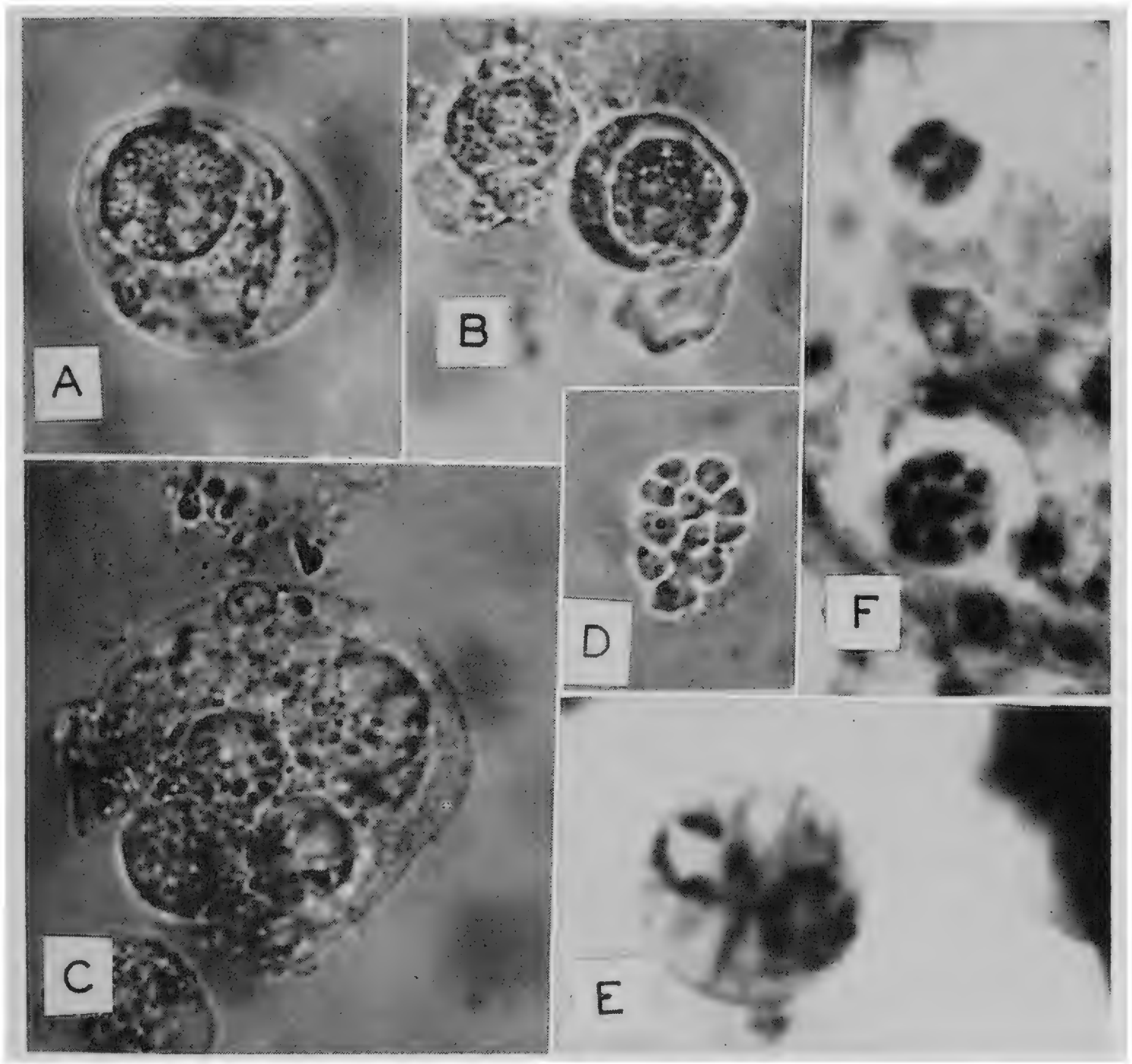

FIG. 1. *Dermocystidium marinum* cells in oyster leucocytes from live smears. A, B. A multinucleate stage prior to segmentation. C. Several hypnospores and one multinucleate stage of *D. marinum* in live smear of the pericardial fluid of a gaping oyster. The broken wall-like structure around the hypnospores is the remains of an oyster leucocyte. D. Daughter cells resulting from segmentation of a plasmodium of *D. marinum*. The plasmodium was gently pressured under a cover glass until the cells fell apart. The dark spots are lipoidal granules. E. A two cell stage with nucleus in mitosis. F. Multinucleate "plasmodium" of *D. marinum* as seen in stained section of the gut epithelium of the host oyster.

"sporangium." They may infect other host cells or grow to become hypnospores (Fig. 2 P).

Steps in Development. Certain features of the early cleavage of *D. marinum* should be mentioned. Very often the thallus will enlarge or grow to near the size of the mature sporangium before division is initiated. The nucleus of this enlarged cell is eccentric, always flattened against the surface membrane of the cell (Fig. 3 B). The nucleus appears to divide into irregular parts by constriction. The daughter nuclei migrate apart and come to rest in the cell at opposite ends flattened against the cell wall. A cleavage plane appears between the two nuclei and the cell divides (Fig. 3 D). Often no cleavage plane is visible, and multiple nuclear divisions appear to take place in a syncytium (Fig. 3 E). When numerous cells result, the whole structure appears to be a multi-nucleate plasmodium. However, careful study indicates that cleavage actually

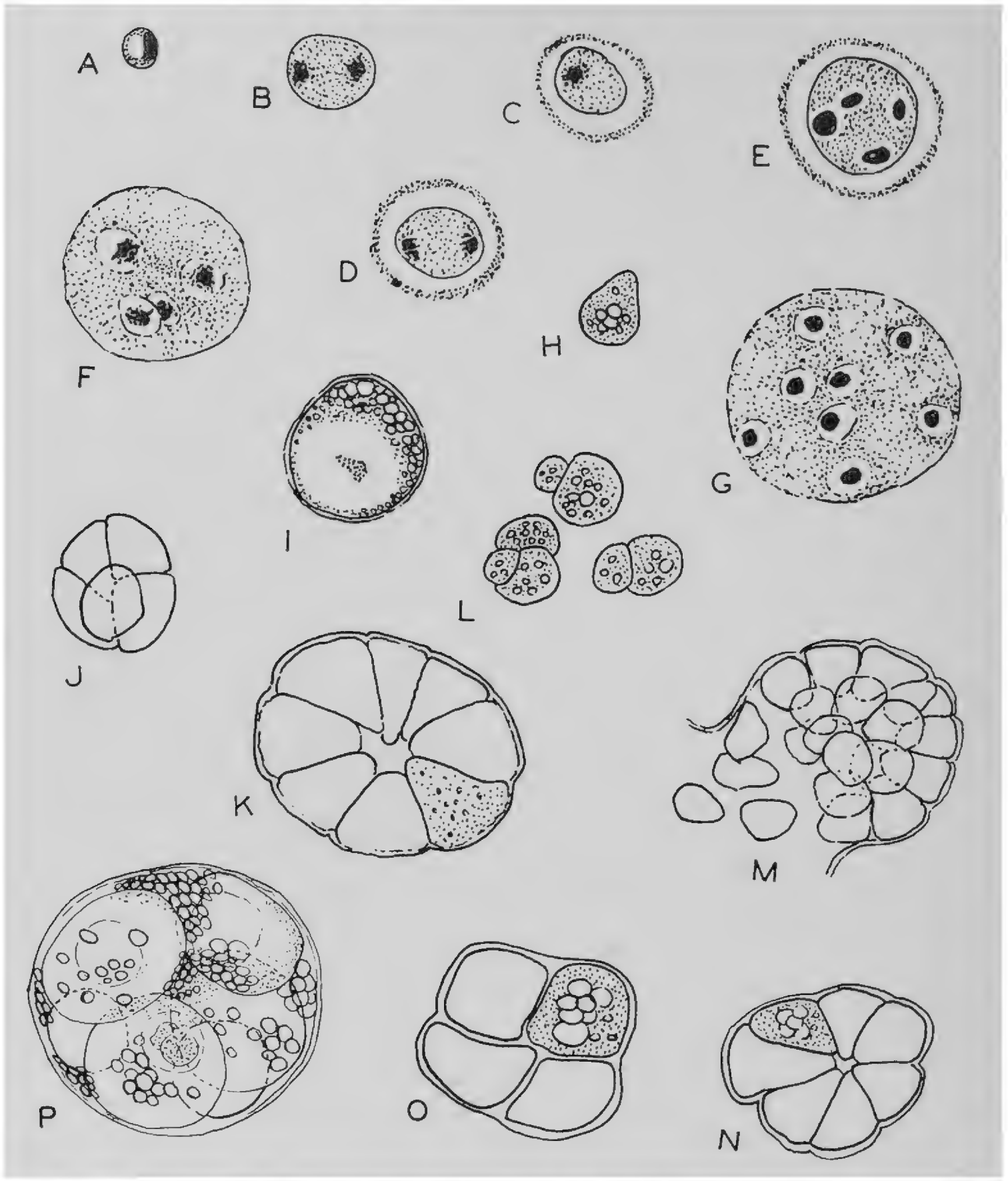

FIG. 2. A small (2 to 3μ) uninucleate thallus from sea water culture. C. With resistant wall. B, D. Binucleate stages, showing late telophase of mitosis. D. With a secreted resistant wall. E, F. Four-nucleate stages, with and without resistant walls. G. Eight-cell stage. H. Sketch of a wedge shaped thallus from a reproducing group; live, unstained, showing lipoid granules. I. Live hypnospore stage with large vacuole from a pericardial smear of a gaping oyster. J. Early cleavage stage as seen in live smear from pericardium of infected oyster. K, N, O. Early cleavage stages from pericardial smear as seen in optical section. L. Budding cells of *D. marinum* from a live smear. M. A mature "sporangium," with a definite limiting membrane, liberating daughter cells (spores?). P. A host cell containing several mature hypnospores, as seen in live smears.

occurs although the planes may not be seen even in well stained preparations. It is believed that growth pressure obliterates the planes of cytoplasmic cleavage.

Ordinarily the two nuclei resulting from the first division do not synchronize their next division. One nucleus divides first resulting in a 3-celled structure (Fig. 3 F). The

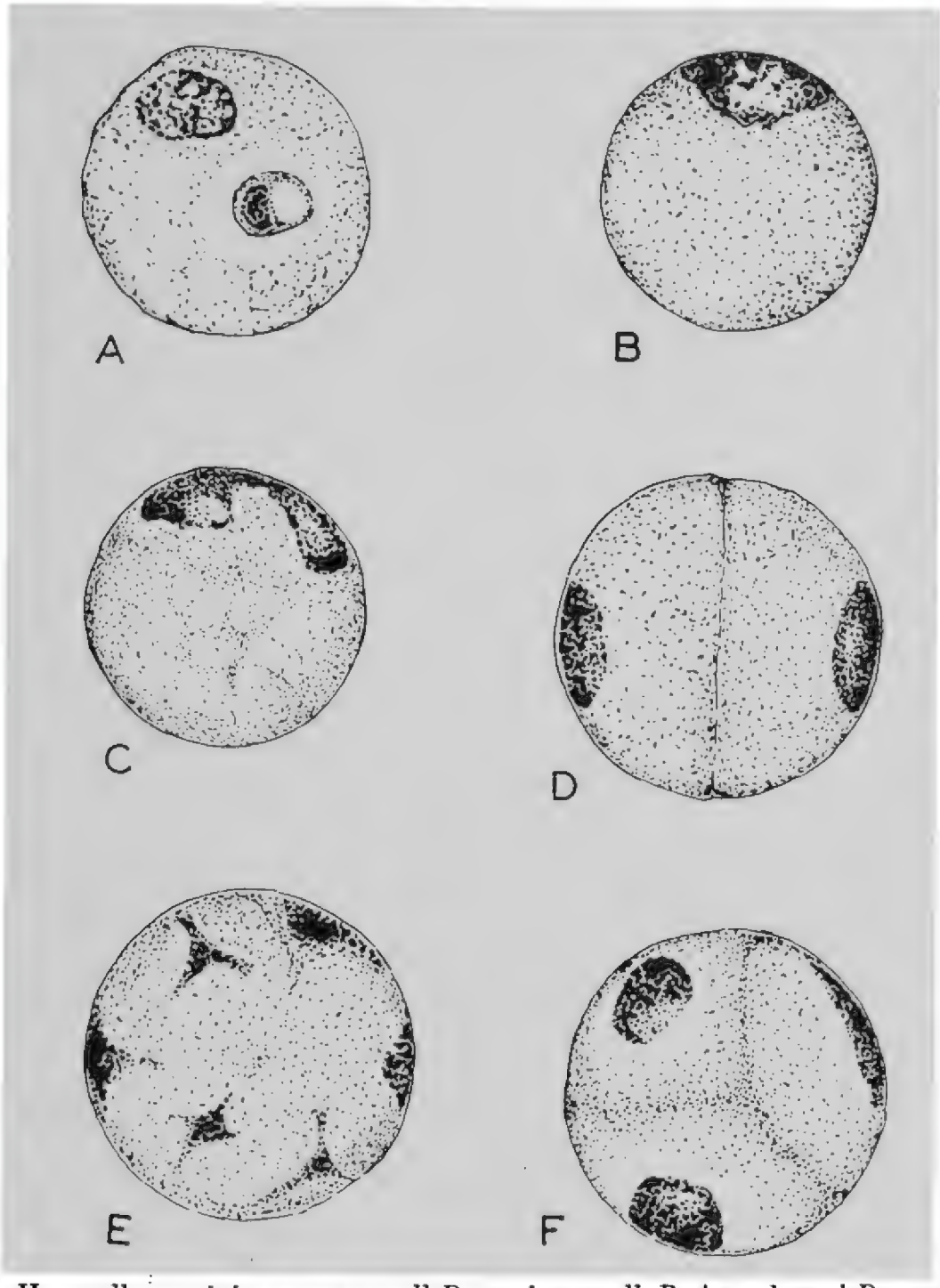

FIG. 3. A. Host cell containing a very small *D. marinum* cell. B. An enlarged *D. marinum* cell with the nucleus elongate and pressed against the cell membrane. C. *D. marinum* cell with nucleus appearing to squeeze apart. D. Two-cell stage with nuclei as far apart in the daughter cells as possible. E. Multinucleate stage with irregular shaped nuclei. F. Three-cell stage of *D. marinum*. One cell is initiating the elongation of the nucleus prior to division.

other then divides, but its spindle is at an angle of approximately ninety degrees to that of the first division. Consequently the cleavage plane is at right angles to the first cleavage plane and to that of the first nucleus to divide. Subsequent cleavage planes are all radially arranged and planes tangential to the surface never occur.

Mitoses are always irregular and the nuclei in process of division appear to be merely elongated or triangular in form without definite membranes (Fig. 3 E). In the early divisions, no karyosome is present and the nucleus appears to be flattened against the cell wall. Many "anucleate" stages may be observed. When rapid division is taking place the chromatin appears to stain very lightly, so that the plasmodium appears to be anucleate. Another artifact frequently observed is a multinucleate stage in which there are numerous deeply-staining granules of volutin-like substance distributed in the "plasmodium." When the actual nuclei stain only very lightly, the volutin granules appear to be nuclei. These granules are probably the "chromidial" nuclei reported for

some of the protistan groups, and apparently all species now assigned to the genus *Dermocystidium* possess them.

Development of the Pre-hypnospore. Cells of *D. marinum* resulting from cleavage (the wedge-shaped thalli) may either repeat the developmental cycle or develop into the vacuolated pre-hypnospores. In sections of heavily infected oyster tissues (Fig. 4 A) the pre-hypnospores are recognized by the large vacuole and the marginal, strongly basophilic cytoplasm. Cells still within the multinucleate masses may develop a vacuole prior to separation of the cells after cleavage. Vacuolate cells tend to grow to a large size. The volutin-like material develops in the vacuole. The volutin granules may often be observed in typical Brownian movement within the vacuole and in all ways appear to be the counterpart of the same material observed in the vacuole of yeasts and other groups of the higher fungi. The staining reaction, however, seems to be much stronger with hematoxylin dyes and resembles the same substance in the vacuole of *Blastocystis.* In addition to the vacuolar inclusion, a viscous fluid accumulates in the vacuole which may be observed in living cells, but apparently is dissolved in fixed materials. The volutin granules may take any shape, but are most commonly nearly spherical and when stained appear to be hollow or composed of concentric layers. Fig. 5 shows

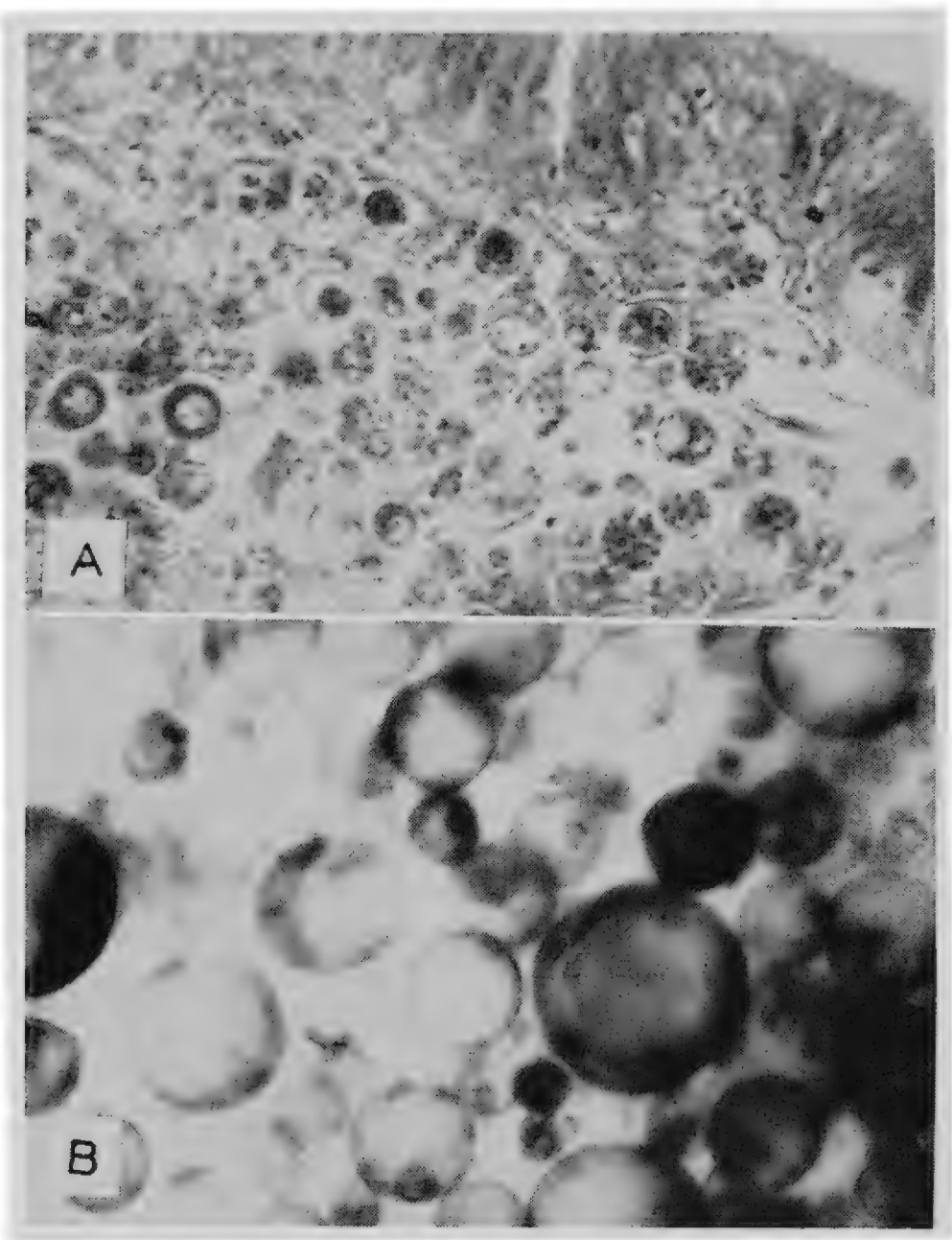

FIG. 4. A. Section of oyster tissue containing various stages in the development of *D. marinum.* B. Cells of *D. marinum* from tissue cultured in fluid thioglycollate medium. These cells were treated with Lugol's solution which produces a blue color in the cell wall. The largest cell is about 75μ in diameter. Photograph of a temporary wet mount.

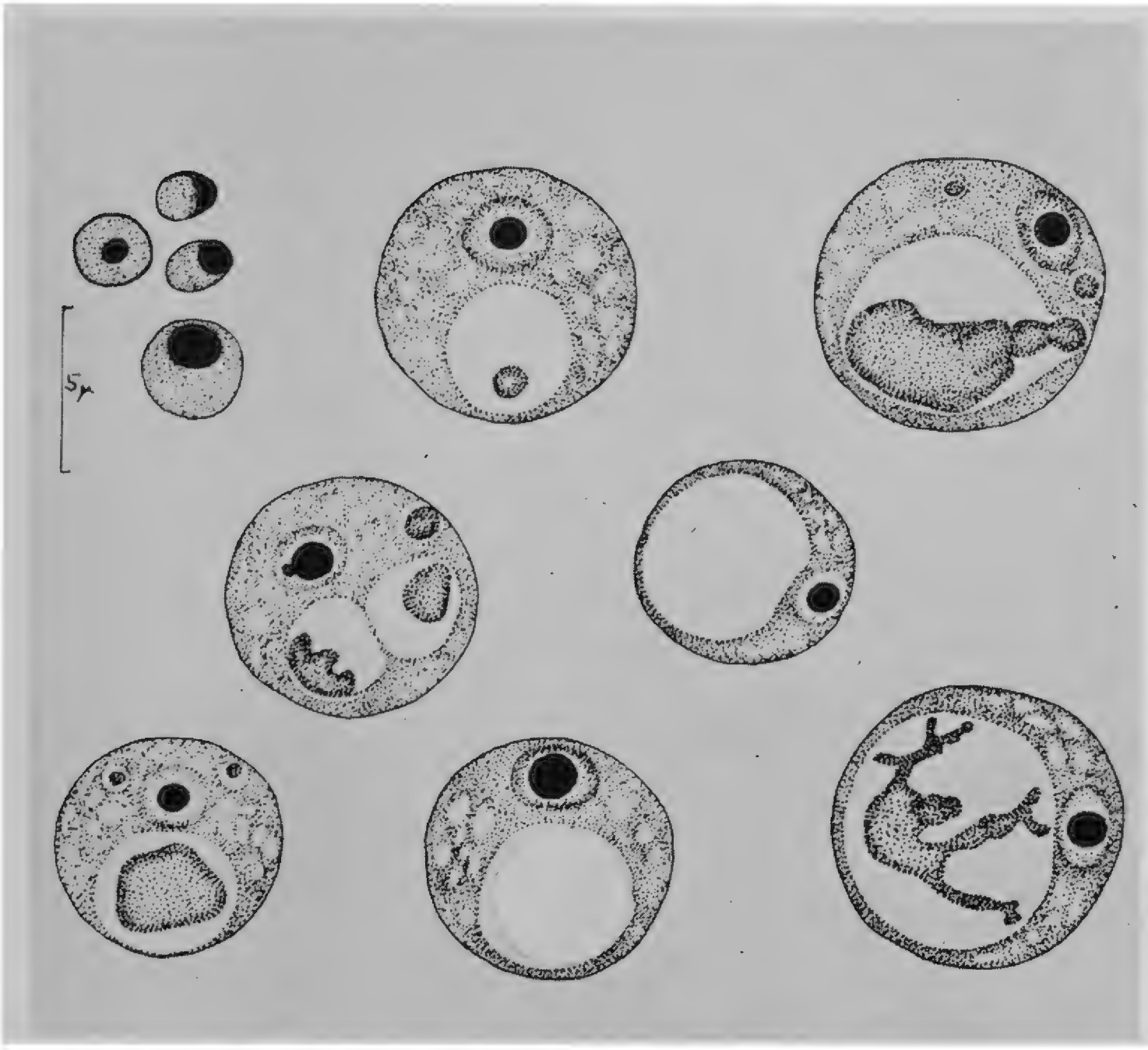

FIG. 5. Free thalli (four small cells) and various forms of developing hypnospores of *D. marinum* as sketched from stained sections of diseased oysters. The inclusions in the vacuoles of the hypnospores are probably of the nature of volutin.

several typical pre-hypnospores as seen in stained sections of infected oyster tissues.

The nucleus of the pre-hypnospores is eccentric, in a "resting" condition. A large karyosome is characteristic of these nuclei, and chromatin material other than the karyosome is pushed to the periphery of the nucleus. It is believed that most of the large pre-hypnospores found in diseased oyster tissue are inactive, especially in cool periods. They appear to await liberation through death of the host or by diapedesis. However, warm periods in winter may stimulate the pre-hypnospores to renewed reproductive activity. It has been observed that few of the pre-hypnospores can be found in recently infected oysters in early summer, but in Louisiana numbers increase through the summer and into the winter.

Mature hypnospores are characterized by a thickened wall, increased size, and increased relative size of the vacuole with its contained nutrient material. Rarely seen in oyster tissues, they probably develop in seawater under certain unknown conditions and serve as overwintering spores (or pre-sporangia ?).

D. marinum Cells in Thioglycollate Culture. Ray (1952a, 1952b, 1954a) discovered that when cells of *D. marinum* were placed in certain nutrient media containing yeast

extract and dextrose, enlargement of the cells took place very rapidly. Cells beginning at about 10 microns may reach 70 microns in size in two to three days incubation. Such enlarged cells have thickened walls which turn blue when treated with Lugol's solution (Fig. 4B). Uncultured cells do not turn blue when treated with iodine solutions. The thioglycollate culture method has been extensively adapted for diagnosis of this fungus disease.

Occasionally cells of *D. marinum* when "cultured" in thioglycollate medium produce elongate tube-like structures growing out of the enlarged cells. These have been interpreted as abortive mycelia. It is possible (1) that they are abortive exit tubes like those which form on mature sporangia in certain chytrid groups, (2) that they are enlargements of budding cells, or (3) that they are structures of a different species of *Dermocystidium*.

Steps in enlargement of cells when cultured in thioglycollate medium are illustrated in Fig. 6. Cells of an eight-cell stage were dissociated by pressure and photographed (Fig. 6A). Subsequently photomicrographs were made of steps in enlargement up to about 24 hours (Fig. 6D). At that time the cells had increased in diameter by about eight times and had laid down a resistant wall. Each cell appears to be one large vacuole. However, a thin layer of cytoplasm rings the interior of the wall. The nucleus enlarges with the cell and can be clearly seen without staining, pressed against the wall as usual.

Under stimulus of nutrient media, all cells of *D. marinum* quickly develop into pre-

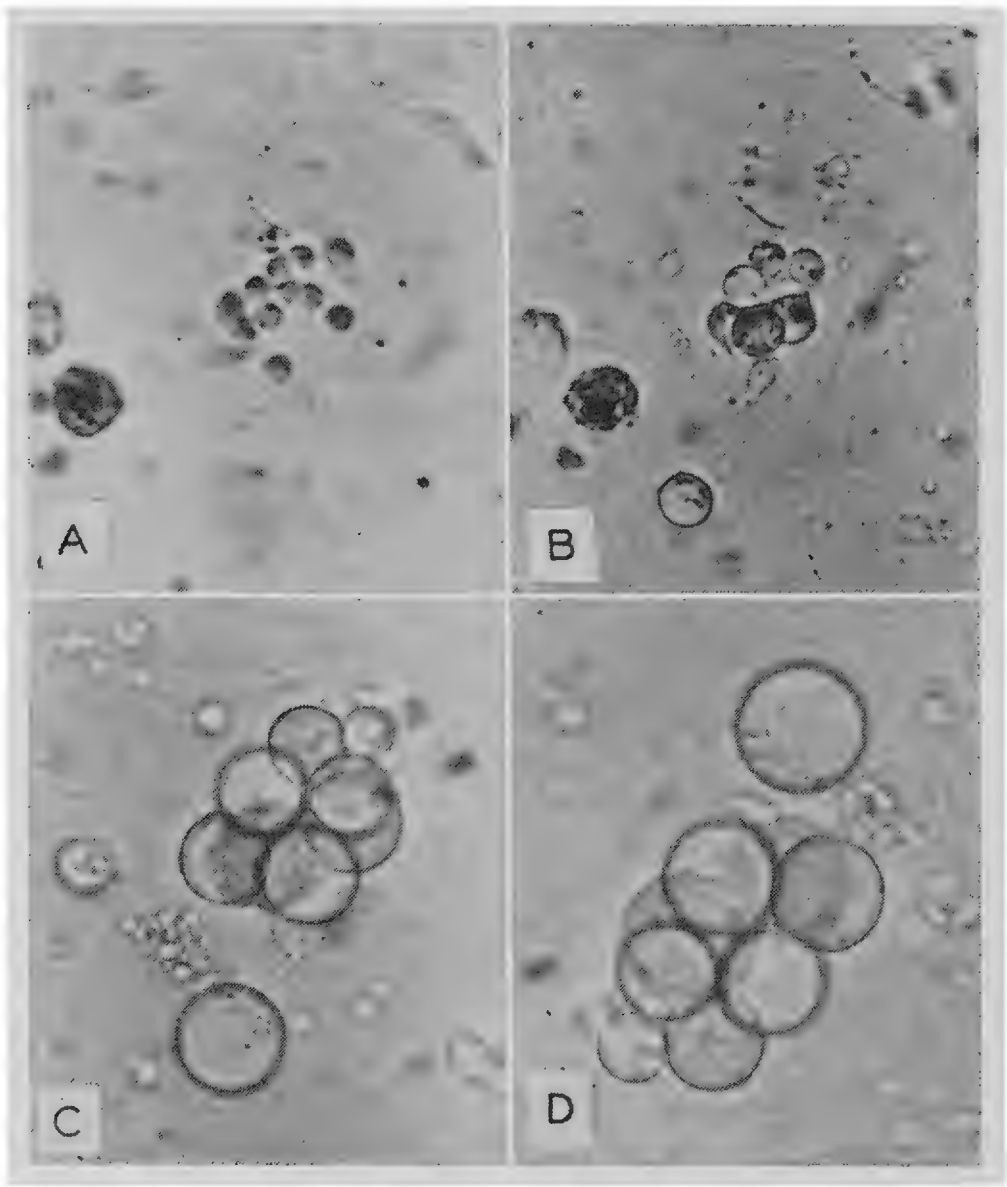

FIG. 6. Enlargement of *D. marinum* cells in fluid thioglycollate medium. A. Disassociated cells of a multicellular stage. B, C, and D. Steps in enlargement up to 24 hours.

hypnospores and then into the hypnospores. Irrespective of the stage of development, this happens to all cells. Stein and Mackin (1957b) analysed the effect of thioglycollate in producing enlargement. Increase in diameter at eight hours was about 50 percent, and at 16 to 18 hours was 100 percent. Those cells which were already in pre-hypnospore stages responded first, followed by other stages. Long experience with cultures indicates that many variables modify the rates of enlargement, and that accurate prediction of the cell size is not yet possible. Some cells develop into enormously large, balloon-like structures more than 100 microns in diameter while others in the same culture may reach only a very moderate enlargement.

It is believed that enlargement of *D. marinum* cells is a normal phenomenon, but that the extreme enlargement observed in concentrated nutrients is abnormal. The author has induced enlargement in seawater, as has Ray (1954a). Owen, according to Ray (1954b), found enlarged hypnospores in four gaping oysters, and Ray also observed the same phenomenon. The author found that *D. marinum* cells kept in sterilized oyster serum, regularly enlarged and were stained blue with Lugol's solution. Enlargement was neither as fast nor as great as in thioglycollate medium or in other media containing the yeast extract or dextrose that Ray and Chandler (1955) thought to be necessary to produce enlargement. Enlargement in oyster serum may rarely be accompanied by cytoplasmic cleavage, and occasionally by budding (Fig. 7). The cleavage suggests that the enlarged cells were proceeding to develop into sporangia. There may be an inhibitor in living oyster tissue that prevents enlargement of the hypnospore or development of a resistant wall prior to discharge into the water.

Studies of *Dermocystidium sp.* in *Macoma balthica* tend to confirm enlargement as a normal phenomenon. Through the courtesy of Dr. Jay D. Andrews of the Virginia Institute of Marine Science (Virginia Fisheries Laboratory), a number of specimens of this mollusc were sent to me at Grand Isle for study. It was found that when a host *Macoma* died, the *Dermocystidium* cells enlarged in the disintegrating tissues just as if they had been cultured in thioglycollate medium. Moreover, the walls of these enlarged cells turned blue when treated with iodine. Microscopic studies also indicated that the cytoplasm of the enlarged cells segmented to produce a number of small cells, assumed to be the equivalent of spores. The hypnospore therefore seems to be a resistant spore or presporangium, which undergoes development only after liberation from the host.

D. marinum in Crude Seawater Culture. Prokop (1950) attempted to culture *D. marinum*. He used many media, including sterile excised oyster tissues, various mycological media, media for protozoan culture, and various combinations. No cultures were successful. However, he established that bacteria were not a part of the *Dermocystidium* syndrome of disease. Excised tissues, removed aseptically, failed to develop bacteria over considerable periods, although they contained numerous *Dermocystidium* cells. These latter failed to reproduce, but were abundant in some preparations. Ray (1952) also made attempts to culture *D. marinum*. These attempts also failed to produce reproduction of the parasites on artificial media.

Both Prokop's attempts and those of Ray suggested that *D. marinum* did not belong to any group of fungi commonly cultured on artificial media. There was an early opinion that *D. marinum* probably was related to *Blastomyces* or *Coccidioides* because of similarities in disease syndrome. The culture failures indicated that this relationship was probably not correct. A study of the characters of *D. marinum* indicated that if it was

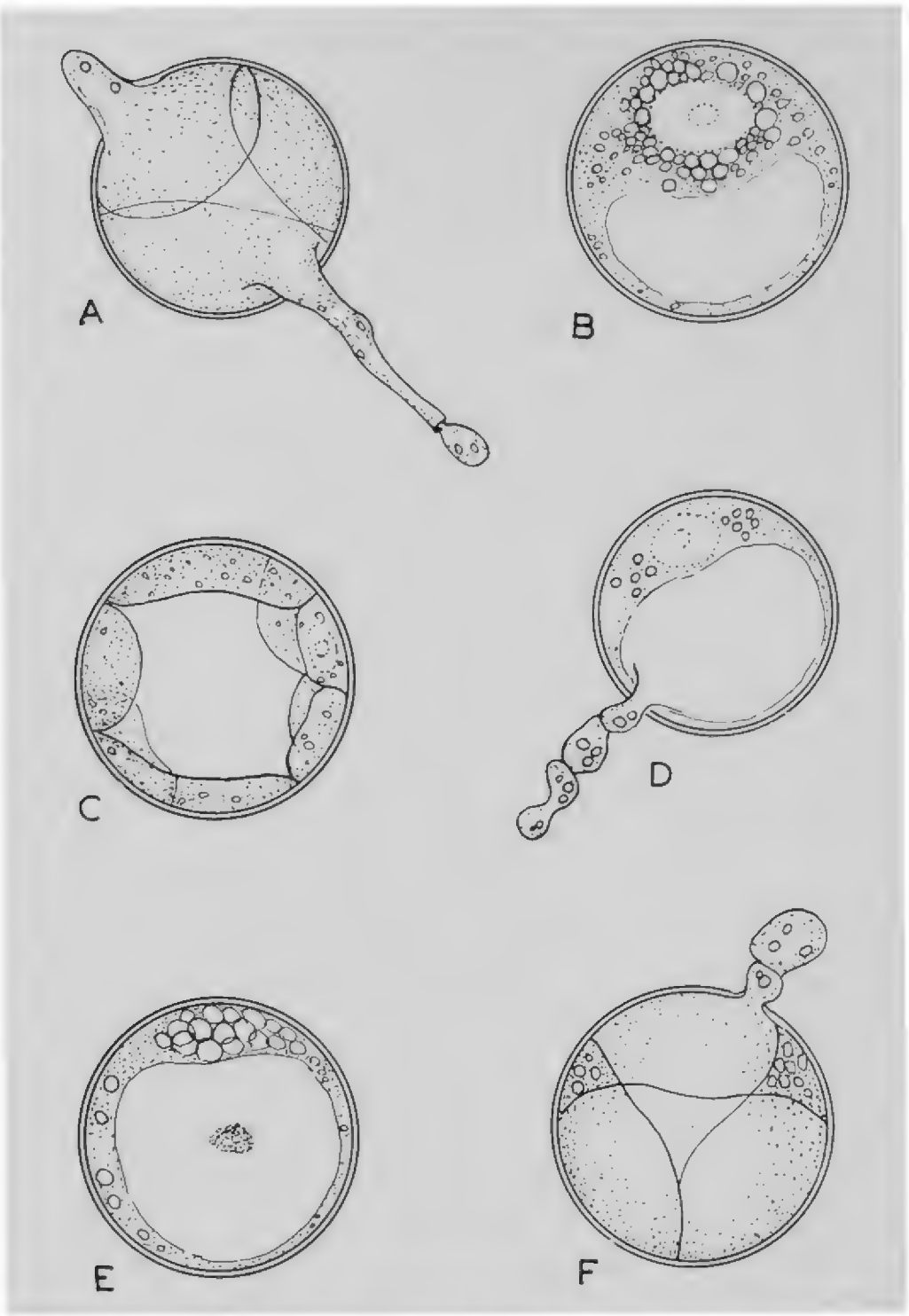

FIG. 7. Sketches of enlarged *D. marinum* cells in oyster serum, unstained, 25 to 40μ. A, D, F. Rare, enlarged cells produce tube-like processes from which small cells may be detached. A, C, F. Cleavage of the cytoplasm. B, E. Enlarged cells without modification except enlargement, and development of fatty granules.

not a yeast-like component of one of the higher Phycomycetes or Ascomycetes, it must belong to one of the fungal groups in which mycelia are wholly lacking. This concept greatly narrowed the field of possibilities so far as recognized groups are concerned.

Based on the work by Prokop and Ray, it seemed probable that an excess of nutrient material was being applied in attempts to culture. Only if *D. marinum* was one of the higher fungi would large concentrations of nutrient be needed. The first step in attempts to culture were therefore made with fluid thioglycollate medium in various dilutions down to 1/100 normal strength. The first trial was successful in producing some reproduction. The data from this attempt are condensed in Table 1. The methods were as follows:

(1) Normal strength fluid thioglycollate medium required 29.5 grams per liter of nutrient material. This concentration in Table 1 is termed a 1.0 concentration.

TABLE 1

Results of attempts to culture *D. marinum* using various dilutions of the standard fluid thioglycollate medium. NaCl was added to these cultures to produce about a 20 ppt medium, and penicillin and streptomycin were added to all cultures

Trial	Concentration of fluid thioglycollate medium						
	1	1/2	1/5	1/10	1/25	1/50	1/100
1	All cells enlarged	All cells enlarged	Enlarged; contaminated	No *D. marinum* cells found	Few reproducing *D. marinum*; contaminated	Moderate no. of reproducing *D. marinum*; contaminated	Rare reproducing *D. marinum* cells found contaminated
2	All cells enlarged	All cells enlarged	No *D. marinum* cells found	No *D. marinum* cells found	Reproduction of *D. marinum* heavy	Reproduction of *D. marinum* heavy	Reproduction heavy; contaminated
3	All cells enlarged	All cells enlarged; not contaminated	No *D. marinum* cells found; contaminated	No *D. marinum* cells found; contaminated	Moderate no. of reproducing *D. marinum*; contaminated	Reproduction heavy; contaminated	Few reproducing *D. marinum*; contaminated
4	All cells enlarged	All cells enlarged; not contaminated	Enlarged	Rare reproducing *D. marinum* cells; contaminated	Moderate no. of reproducing *D. marinum*; contaminated	Moderate no. of reproducing *D. marinum*; contaminated	No *D. marinum* cells found; contaminated
5	All cells enlarged	All cells enlarged; all dead	No *D. marinum* cells found	No *D. marinum* cells found	Few reproducing *D. marinum*	Few reproducing cells found; contaminated	No *D. marinum* cells found; contaminated

(2) The effect of various concentrations of potassium iodide was also tested in this series. Since no effect was demonstrated, no data are reported.

(3) Volumes of fluid thioglycollate medium, K I, antibiotics (pencillin and streptomycin) and homogenate of infected oyster tissue were so mixed to produce 10 cc of inoculated culture medium per screw cap tube.

(4) Tissue from a gaping oyster was carefully broken up in a Waring Blendor. The resulting homogenate was strained through multiple thicknesses of cheese cloth to remove excess tissue. Counts of *D. marinum* cells were made from 10 aliquots of this homogenate and averaged. Counts were made with a standard haemocytometer. The homogenate was then diluted to contain approximately 200,000 *D. marinum* cells per cc of homogenate. One cc of the homogenate was introduced into each of 35 screw cap culture tubes which contained various concentrations of fluid thioglycollate medium as shown in Table 1.

(5) Culture tubes were kept at "room temperature," 25° to 30°C, approximately.

(6) Study was begun on July 6, 1954, and the tubes were examined on July 13, 1954 and subsequently on July 17, 1954 when the study was completed.

The data from this study (Table 1) showed that enlargement did not occur in dilutions of 1/10 to 1/100 strength. One culture of 1/10 strength showed a few reproducing *D. marinum,* but no *D. marinum* cells were recovered in four cultures. In all 1/25 and 1/50 strength cultures reproduction took place, and good cultures developed in a few tubes. Only one of the 1/100 strength cultures was successful, this one producing very good reproduction. Overall the 1/50 strength cultures were rated best with two fair and two very good reproducing tubes.

Most of the cultures produced in dilute fluid thioglycollate medium were contaminated, either with bacteria or with filamentous fungi, mostly *Fusarium.* In those cultures in which bacteria developed *D. marinum* failed to reproduce, although enlargement occurred under conditions of bacterial contamination. When fungi were contaminants they appeared to have no effect on reproduction.

All cultures were dead or moribund eleven days after initiation. Examination showed that the fungus cells in 1/25 to 1/100 dilutions, without bacteria, reproduced more or less rapidly for about a week and then died. However, any reproduction at all was a step forward and was regarded as having bolstered the theory that dilute media were necessary for reproduction. The fact that a little oyster tissue was introduced into the cultures left in doubt the importance of the diluted medium. It was not known whether or not the cells, during the short period of reproduction, depended for nutrient substance on the fluid thioglycollate medium, the oyster tissue, or stored nutrient in the *D. marinum* cells.

The form of reproduction in these cultures was unexpected. Most cells reproduced by budding and fission into small loosely aggregated clumps of cells (Fig. 8A). Budding in oyster tissue had been observed rarely and was regarded as possibly atypical. In these cultures reproduction was mainly by budding and cleavage. Some advanced hypnospores apparently developed directly into sporangia containing numerous very small aplanospores, which were often liberated by rupture. Occasionally reproduction by means of the radially segmenting, morula-like bodies was observed as commonly seen in oyster tissue.

In cultures of one-half the normal concentration of fluid thioglycollate medium the cells enlarged, but no reproduction occurred. In some of these enlarged cells, cytoplasmic

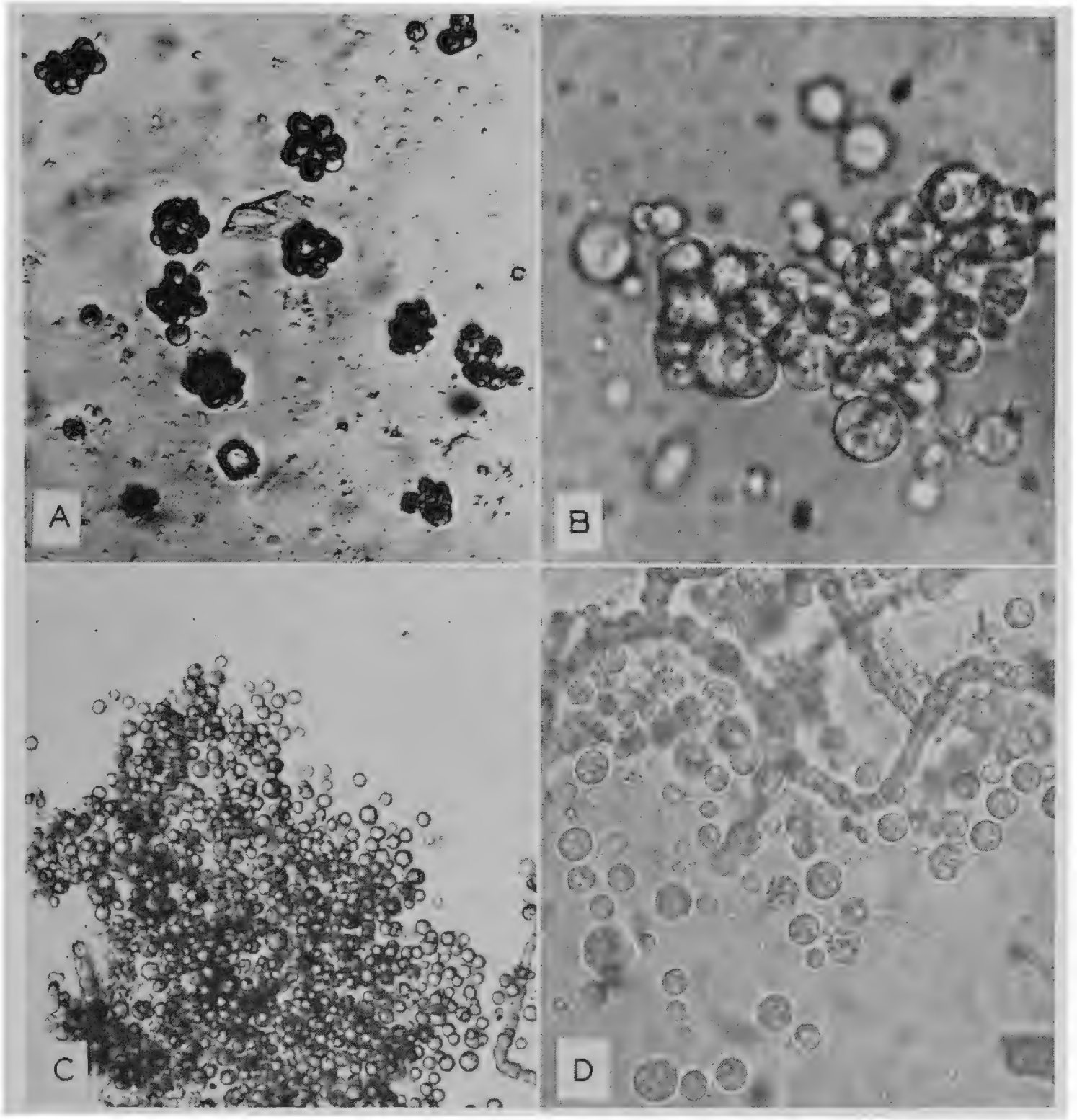

FIG. 8. Reproduction of *D. marinum* in fluid thioglycollate medium diluted to 1/50 strength. A. Fission and budding produces small loosely aggregated clumps of cells. B. Reproduction of *D. marinum* in sterile "sea-water" cultures. The grape-like clusters develop by fission and budding in early stages of culture. C. *D. marinum* in sea-water culture. Most of these "cells" are multinucleate, and in many cases the cleavage planes could be clearly seen. D. *D. marinum* in "sea-water" culture enlarged to show radial cleavage planes in some sporangia (?).

cleavage occurred, but did not continue beyond production of a few cells. No nuclei could be seen in these cells, but the original large eccentric nucleus was clearly seen. It is assumed that, if nuclei existed in the cytoplasmic segments, they were chromidial in origin.

A second culture series which used only 1/40 strength dilutions of the fluid thioglycollate medium, but was otherwise not different from the first series, was a failure. A third series incubated at 15°C also failed. A fourth series was held at 30°C with dilutions all at 1/40 strength with better results. Out of 22 cultures, four were judged to be good, with satisfactory reproduction of *D. marinum*. It was noted that these were cultures which were not visibly contaminated by either yeasts, molds, or bacteria. Series four

was unsatisfactory as a whole, adding little or nothing to the first series. The cultures which were successful ceased reproducing within 10 days after the cultures were set up. Attempts to subculture were without success. Apparently massive inoculation is prerequisite to successful culture.

Following these efforts, a number of attempts to culture were made using oyster serum (sterilized by filtration), yeast extract, peptone, and fluid thioglycollate medium, in various combinations and singly. All of these, including oyster serum, produced enlargement of *D. marinum* cells. Reproduction failed to occur except, rarely, by budding and by cleavages.

Culture with Fusarium. On November 2, 1954, a gaping oyster was minced with a Waring Blender, and the *D. marinum* cells were separated from the tissue by straining through several thicknesses of cheese cloth. Most *D. marinum* cells wash through such a strainer, while most tissue debris is eliminated. Cells thus recovered were added to vials of sterile seawater in which were placed excised bits of oyster mantle in an attempt to infect these tissues. After the mantle tissues were removed, the vials, containing large numbers of *D. marinum* cells in sterile seawater fortified with streptomycin and penicillin, were allowed to stand unmoved for a period of 15 days. On November 17, 1954, these vials were examined. All had developed a heavy mycelial mat found to be a species of *Fusarium.* A considerable part of the seawater had evaporated. Examination of the *Fusarium* showed that in all vials (14) masses of *D. marinum* had developed. No nutrient had been added to the seawater (sterilized by filtration). The homogenized oyster tissue and the dissolved substances in the seawater supplied the only nutrient available. Besides *Fusarium*, there were several other contaminating species of mycelial fungi, ciliates, and yeasts in some of the cultures.

The *D. marinum* cells had reproduced via the radial cleavage route, as in the oyster, and the cultures when examined were made up mostly of sporangia or the aplanospores which came from them. At the time of examination no oyster tissue could be found in the cultures, but most of the *D. marinum* cells appeared to be alive. Attempts to subculture from these seawater cultures failed.

New cultures were set up on November 20, 1954, using sterile seawater fortified with antibiotics. As much oyster tissue as possible was strained out. Cells and sera were diluted to 1/200 with sterile seawater. Two and one half cc of this dilution was added to each of 20 penicillin bottles containing sterile seawater. Some of these cultures were inoculated with *Fusarium* spores from pure culture of the latter; others were inoculated with *Fusarium* mycelia from previous cultures, and several vials were inoculated with spores from a *Candida* culture. One set of vials was left without *Fusarium* or *Candida.* Each of these groups contained five cultures.

At the end of one day, reproduction in some tubes had definitely begun. Six satisfactory cultures were achieved out of the 20 attempts. These six were still in good condition on December 9, 1954 (after 19 days). All cultures, irrespective of treatment, became dominated by *Fusarium.* Two of the cultures produced great masses of thousands of *D. marinum* cells and sporangia (Fig. 8C). At 19 days nearly all were multinucleate sporangia, as shown by stained smears. Nearly all reproduction was by radial cleavage (Fig. 2: J,K), but some budding and fission in early stages was observed.

Some of these cultures produced giant sori of sporangia, the first of these seen (Fig. 9D). Some of these were up to 40 microns in diameter and had very thin walls.

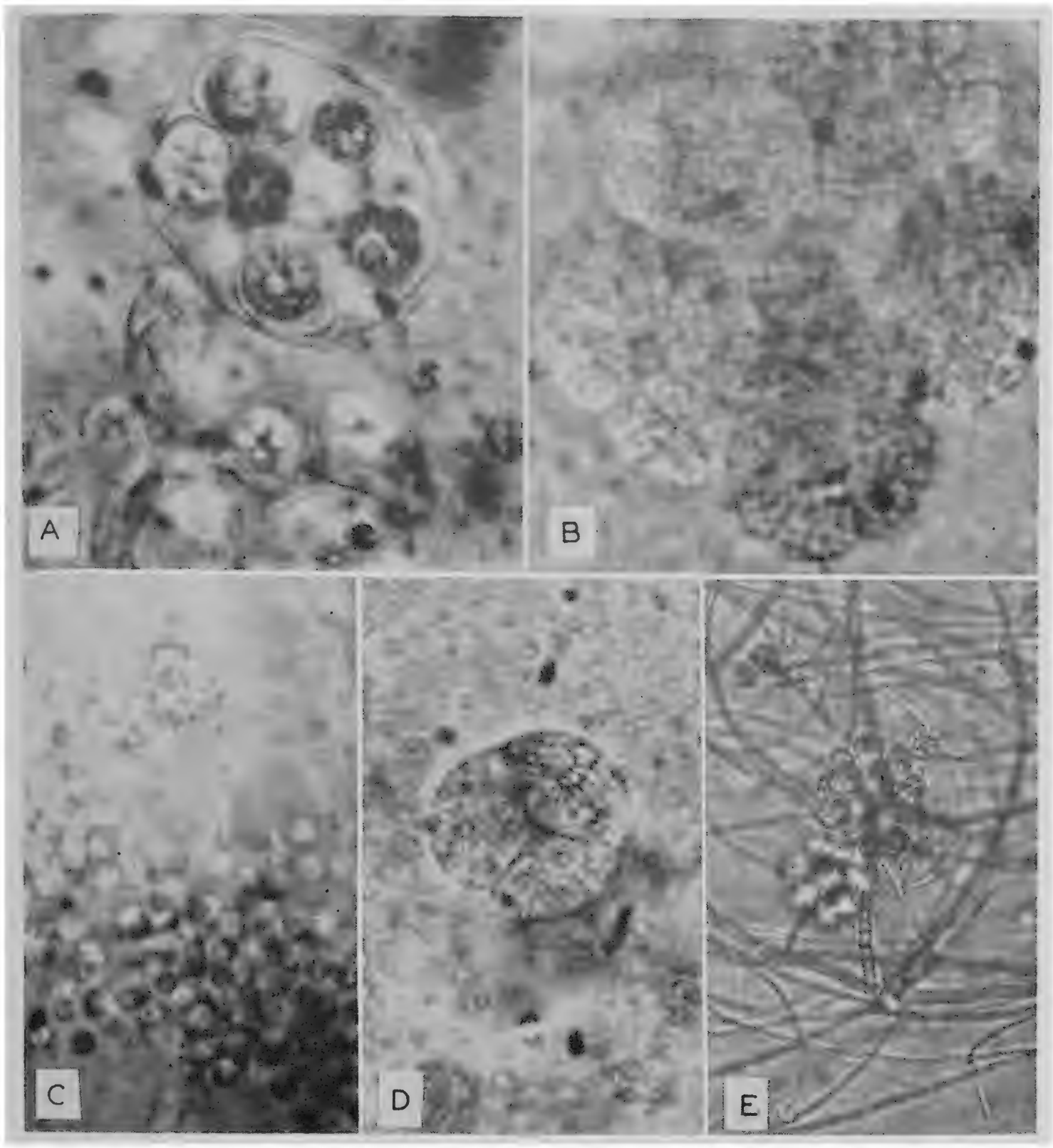

FIG. 9. Sori and aplanospores of *D. marinum* in sea-water culture. A. A sorus containing several sporangia (?); B. Mature sorus containing aplanospores. C. Aplanospores from a sorus as in B. D. An early thin-walled "giant" sorus of four multinucleate sporangia (3 shown in the photograph). E. Sori of sporangia with thin walls.

Each "cell" was multinucleate and was a sporangium. These giant sori appeared more or less consistently in cultures and often developed numerous sporangia.

Studies to determine whether or not introduction of glycogen into cultures was helpful were carried out with negative results. Numerous cultures with graded amounts of glycogen added were less successful than were controls using only seawater. Nutrient agar failed to improve the culture methods. The final technique that gave the most consistent results was as follows:

1. Culturing was done in 100 mm covered petri dishes to permit maximum aeration of a thin layer of sterile seawater on the bottom of the dishes.

2. *D. marinum* cells were introduced into the sterile seawater in small 3 to 5 mm-square pieces of heavily infected gill tissue. Cultures were made only from gaping oysters with strong ciliary action and without sign of bacterial degeneration.

3. Gill tissues were washed in successive baths of sterile seawater to remove as many bacteria, fungus spores, flagellate and ciliate protozoa, and amoebae as possible from the gill net.

4. Every effort was made to control bacteria, apparently the cause of most culture failures. Penicillin and streptomycin were used in wash waters and cultures.

5. Each small block of gill tissue containing *D. marinum* cells was inoculated with spores of *Fusarium* from pure culture. *Fusarium* grew well in the still-living gill tissue, and its mycelia, in about a week's time, made a solid mat penetrating the tissues and subsisting on them. In a week to ten days the host tissues were absorbed. *D. marinum* cells continued development in the oyster tissue as long as it lasted. When the host tissues were gone, the *D. marinum* cells continued development in the mycelial mat, attached to the strands. It is believed that the *Fusarium* produced an antibiotic protective against bacteria, and that it may also have produced some growth-promoting hormone effective on *D. marinum*.

6. Cleansing of glassware was done without detergents, and metal instruments were avoided.

7. Cultures were kept in a constant temperature box at 23°C.

In general, when *D. marinum* cells are free of host tissue, the first reproduction takes place by fission and budding, resulting in grape-like clusters of cells (Fig. 8B). Reversion to radial cleavage takes place early, and most subsequent reproduction duplicates the steps in the oyster tissue but with some differences. First, the cells in many cases secrete resistant walls, and second, the size of the mature sporangia is definitely much larger than those in oyster tissue. They may reach a diameter of more than 20 microns (Fig. 8D). Nuclear multiplication is always quite regular (2, 4, 8, 16 cells, etc.), and takes place without cytoplasmic cleavage up to the final divisions which separate the daughter cells. Cells resulting from such division immediately repeat the cycle, but not indefinitely. Apparently there is an end stage, which is an aplanospore (Fig. 9C).

Sori developed in some cultures (Fig. 9: A,D,E). These produce aplanospores, which may develop to new sporangia prior to break-up of the old sporangial wall (Fig. 9A). Such sori may be up to 40 microns in diameter. Some of the sori are formed from enlarged uninucleate cells with very thin walls. The cleavage pattern of these is always identical to that of their smaller counterparts, but the result is a sorus containing many more aplanospores (Fig. 9B). The role of these giant sori is difficult to assess. It is not known whether they represent responses to abnormal conditions, or whether they are normal themselves, and appear only under very favorable conditions.

These large sori mature as a spore mass which looks much like similar structures in the Plasmodiophorales (as in *Spongospora*) (Fig. 9B). However, no flagellated zoospores have been observed to be liberated from these masses. It is possible that they are abnormal.

Uniflagellate and biflagellate organisms commonly appeared in many cultures. Because their appearance was not consistent, it is believed that these, with one possible exception, were zooflagellates and not flagellated zoospores. The exception was a zoospore with single trailing flagellum which in several cultures attacked ciliate protozoa. In some cases the attacks produced chytrid epidemics which wiped out the ciliate popula-

tions. The nature of these parasites is unknown, except that they obviously belong in the Chytridiales. It is believed that flagellated zoospores of *D. marinum* might attack any available cells in a culture. The presence of foreign chytrids in the crude cultures suggests at least the possibility that all stages observed in the cultures may not have been *D. marinum* although most undoubtedly were.

The Genus *Dermocystidium* Perez, *D. marinum* and Related Low Fungi

Perez (1907) erected the genus *Dermocystis* and later (1908) amended it to *Dermocystidium*, because of pre-occupation. In 1913 he gave an extended description of the type species, *D. pusula*. It is unfortunate that *D. pusula* still is known only from the mature spore (hypnospore) stage with its large eccentric nucleus and vacuole with inclusion body. However, several other species of the genus have been described and from these a more complete picture of the genus is available. All species now placed in *Dermocystidium* (excepting *D. marinum*) are parasites of the skin, gills, or muscles of amphibians and fishes. One of the unsolved problems is the relation of *Dermocystidium marinum* to the complex of species parasitic in vertebrates. A second problem is that of the taxonomic relationship of *D. marinum* within the lower Phycomycetes. These two problems are not as yet resolvable. That is not because of lack of information concerning *D. marinum*, but because (1) the *Dermocystidium* group parasitic in vertebrates is incompletely known, and (2) the large group of genera which, like *D. marinum*, belong vaguely near the lower Phycomycetes, are themselves known mostly from fragmentary accounts. The group of genera referred to, and discussed below, are taxonomic orphans in the sense that they are generally excluded from, or included only doubtfully in, known and accepted taxa. They have been shifted by various authors from the protozoa to the lower fungi or vice versa, or occupy a vague intermediate position on the borderline between the two groups. *D. marinum* and the complex of related species shown by Andrews (1955) and Ray (1954b) to exist in marine bivalve molluscs and annelids stand in the middle of these taxonomic orphans. The purpose of this section is to analyze the facts as known and define the problems remaining.

Dermocystidium pusula Perez (1907) parasitizes the skin of *Triton marmoratus* and *T. cristatus*. Only the mature spore (prehypnospore?) stage is known. This stage occurs in small cysts in the skin. These cysts are undivided and appear to contain single thin-walled sporangia, and the "spores" are not infective. Each spore has a large eccentric nucleus, a basophilic cytoplasm containing chromidia, and a large "vacuole" containing an inclusion of the general staining reaction of volutin. The pre-hypnospore stage of *D. marinum* is approximately identical to that of *D. pusula*, hence the placement in the genus *Dermocystidium* (Mackin, Owen and Collier, 1950).

Other species placed in the genus *Dermocystidium* Perez by subsequent authors, and which parasitize fishes and amphibians, are nearly identical to *D. pusula* in the final spore stage, but species of this group vary much in details of development.

Leger (1914) described *Dermocystidium branchialis* from the gills of *Trutta fario*. This author observed developmental stages in small cysts, 0.2 to 0.3 mm in diameter. These developing stages consisted of granular cytoplasm, like a plasmodium, containing scattered and very small nuclei. Scattered in the cytoplasmic mass were large inclusions which stained strongly with basic dyes. As the cysts developed, the cytoplasm of the

multinucleate plasmodia broke up into islets, each with a single nucleus. These islets gave rise to the hypnospores.

D. ranae Guyenot et Naville, 1922, which parasitizes the skin of *Rana temporaria* and *R. esculenta* develops through a granular plasmodial stage as does *D. branchialis*. The cysts of *D. ranae* are U-shaped rather than spherical or oval. The early plasmodium is "anucleate" when tested by the Feulgen method (Broz and Privora, 1952) but contained deep-staining granules (of volutin?) when dyed with ordinary basic stains.

D. vejdovskii Jirovec, 1939, parasitizes the gills of the pike *Esox lucius*. Young cysts of this species also contain only granular plasmodium. In development this plasmodium first breaks into many nucleated sub-plasmodia, then these produce spores. This involves, therefore, the development of a sorus of sporangia rather than a single sporangium, and in this respect this species differs from the others.

Davis (1947) described *D. salmonis* from the gills of the salmon (*Oncorhynchus tshawytscha*). This author failed to find plasmodia in the young cysts but did find immature cells of *D. salmonis* which were reproducing by binary fission. These cells differed from the mature spore stage by the lack of a vacuole with inclusion, and many were binucleate. It seems possible that Davis failed to find the earliest stage, which may have been a plasmodium. Nevertheless the interpolation of a binary fission or budding stage in the developmental cycle is of great interest. The description by Reichenbach-Klinke (1950) of *D. percae* from the skin of the perch (*Perca fluviatilis*) failed to add new data on the life cycle.

Hoshina and Sahara (1950) described *D. koi* from the skin and muscles of the carp (*Cyprinus carpio*). These authors failed to find plasmodial stages, but their description adds an important detail to the life cycle. *D. koi* proliferates in muscle and skin of the host into numerous polyp-like masses of cysts (or sporangia). These resemble very strongly the masses of sporangia characteristic of *Rhinosporidium seeberi*, parasitic in the nasal epithelium of humans and cattle. The proliferations of *D. koi* may result from auto-infection from an original single cyst. If this is so, then the hypnospores of this species are infective, at least under certain unknown conditions. An alternate theory is that the original infective cell, if not isolated by the host, may fragment, the parts then being distributed by phagocytes, each forming a new focus of infection. A characteristic of the cysts and polyps of *D. koi* is a meagre host response resulting in incomplete isolation of the thin-walled sporangia by the cyst wall.

All of the species of *Dermocystidium* are similar in that the hypnospore (1) develops a single large vacuole in the cytoplasm which contains volutin bodies in various forms, (2) has a basophilic-staining reticular cytoplasm which contains nucleus-like chromidia, and (3) has a large eccentric vesicular "resting" nucleus with a large karysome. In all cases the pre-hypnospore has a thin unsculptured wall.

Vacuolated cells of the *Dermocystidium* type occur in other widely separated genera. For example, in the chytrids, such vacuolate hypnospores occur in *Cladochytrium* (Cladochytriaceae). They occur also in the Actinomyxidian, *Sphaeractinomyxon*, and in *Coelomycidium*, commonly placed in the Haplosporidia. The hypnospore of the basic type found in *D. pusula* has characters found in widely separated species of Protozoa and Fungi. The best that can be said for the placement of *D. marinum* in the genus *Dermocystidium* Perez on the basis of the form of the vacuolated pre-hypnospore is that this character indicates that it could be a member of the genus.

The placement in the genus *Dermocystidium* of the various species parasitizing verte-

brates has not been questioned. But all were relegated to the genus *Dermocystidium* on the basis of the similarity of the final spore form just as was *D. marinum*. As summarized above, some of these assumed species of *Dermocystidium* (*D. salmonis*, *D. koi*) differ much from others of the genus in developmental cycle. If *D. marinum* were separated from the genus *Dermocystidium*, there would be ample reason also to separate other species now included. If the form of the pre-hypnospore is not important, *D. marinum* appears to be related to such genera as *Ichthyochytrium* Plehn (1921) or to certain genera of the Haplosporidia or to *Sporomyxa* Leger 1908. All of these have a basophilic cytoplasm containing chromidia, and all develop by progressive cleavage to a subspherical sporangium-like body in which spore-like cells are produced.

There are about thirty of the more or less distantly related orphan genera mentioned above. The taxonomic status of these is in doubt and nearly all have been referred more or less positively to the "chytrids," to the Myxomycetes, or to the Haplosporidia. Occasionally the placement is narrowed and the groups generally mentioned as most nearly related are the Olpidiaceae and the Synchytriaceae. Three genera of these orphans are known to have flagellated zoospores and must belong to some chytrid group. These are (1) *Blastulidium* Perez, 1903, with a uniflagellated zoospore, (2) *Blastulidiopsis* Sigot, 1931, with a biflagellate zoospore, and *Dermomycoides* Granata, 1919, with a uniflagellate zoospore. *Blastulidium*, except for the zoospore, has little resemblance to most chytrid groups, but probably is near to the Synchytriaceae. *Blastulidiopsis* is basically like *Blastulidium*, except for the biflagellate zoospore. Following the mycologists' precept that the number of flagellae is all-important and nothing else bears much weight, this organism most likely would belong to the Plasmodiophoraceae or the Lagenidiales. *Dermomycoides* occurs in cysts of the skin of *Triton palmatus* and these cysts are precisely like those of *Dermocystidium pusula* Perez. However, the flagellated zoospores produced in the cyst are not in any detail like the hypnospores of *Dermocystidium* and the genus has been referred to the Olpidiaceae.

The fact that other genera of the "orphan" group have not been shown to have flagellated zoospores does not mean that they may not have them. One is apt to overlook the effect of parasitization of animal rather than plant hosts. Most parasites of animal hosts have been fixed, sectioned and stained before the parasites are found. Their appearance when viewed alive is quite different from their appearance when stained. *Haplosporidium* when seen alive looks like a low fungus; stained it shows characters similar to those of some Microsporidia.

The data indicate, however, that most of the genera of the "orphan" group have no flagellated zoospore, and thus must be excluded from the known taxa of low chytrids or the Plasmodiophoraceae. That leaves only the alternative of placing them in the Haplosporidian wastebasket, which now contains obvious fungus genera. It is believed that no good purpose is served by continued placement of doubtful genera in the order Haplosporidia. It only defers the time when the group of "orphan" genera are grouped together in a taxonomic unit and become one of the lower chytrid groups. At present it matters little that this group probably would not be a pure phylogenetic line with a single stem. It is generally admitted that most of the lower Phycomycete groups may be polyphyletic.

Included in this group are: *Blastocystis* Alexeieff, 1911; *Rhinosporidium* Wernicke, 1903; *Chytridiopsis* Schneider, 1884; *Chytridioides* Tregouboff, 1913; *Dermomycoides* Granata, 1919; *Lymphocystidium* (Jirovec), 1936; *Ichthyochytrium* Plehn, 1921;

Mucophilus Plehn, 1921; *Hepatosphaera* Gambier, 1924; *Histocystidium* Goodchild, 1953; *Dermosporidium* Carini, 1940; *Mycetosporidium* Leger and Hesse, 1905; *Icthyosporidium* Caullery and Mesnil, 1905; *Coelosporidium* Mesnil and Marchoux, 1911; *Sporomyxa* Leger, 1908; *Bertramia* Caullery and Mesnil, 1899; *Polycarium* Stempell, 1903; *Erythrocytonucleophaga* Ivanic, 1942; *Blastulidium* Perez, 1903; *Endoblastidium* Codreanu, 1931; *Blastulidiopsis* Sigot, 1931; and several others, including *Dermocystidium* Perez, 1908.

Dermocystidium Perez seems to be most closely related to *Dermosporidium* Carini (1940). The only difference between these two genera is that the final spore stage in *Dermosporidium* has several vacuoles with inclusions and a central nucleus, while in *Dermocystidium* the final spore stage has one large vacuole and an eccentric nucleus. *Dermocystidium* is also closely related to *Rhinosporidium*. This latter genus produces spores which are quite similar to those of *Dermosporidium*. Elles (1941) relates *Rhinosporidium* to the Olpidiaceae, as do Chaudhuri, *et al.* (1945).

Dermocystidium marinum fails to agree with the other species of the genus in that the number of spores produced within the sporangium is much smaller and there is no granular plasmodial stage. But the "plasmodial" stage has not been found in all the species assigned to *Dermocystidium*, notably *D. salmonis* Davis and *D. koi* Hoshina and Sahara.

D. marinum grown in seawater culture rarely develops a vacuole with inclusion body. According to the medium or host conditions under which *D. marinum* is grown, the form of the cell may be modified. Many immature *D. marinum* cells in the oyster host do not have a vacuole. The presence or absence of a "plasmodium" in some related genera (*Chytridiopsis*) seems to be dependent on unknown factors. Whether or not a "plasmodium" forms may merely be a matter of whether or not cytoplasmic cleavage parallels nuclear divisions. It seems best under the circumstances to leave *D. marinum* in the originally assigned place. Further study of the complex of species in marine molluscs and annelids which respond to the thioglycollate culture test may indicate that the marine forms should be removed from *Dermocystidium* Perez and either placed in a new genus or assigned to one or another of the genera listed above.

There can be little doubt that *D. marinum* is a fungus. Strong evidence of its fungal nature is furnished by Ray's (1952a) demonstration that the hypnospore wall, after culturing in various media, will react with iodide to produce a deep blue color, and by his published photomicrographs showing tubular growths from the enlarged hypnospores described above. Stein and Mackin (1957a) showed that the wall of *D. marinum* cells in oyster tissue stained deep red with the periodic acid-Schiff reaction, a characteristic of the cell wall of fungus cells. The cell wall of *D. marinum* was shown probably to contain hemicellulose also.

Since mycelia are wholly lacking in the development of *D. marinum* either in the host oyster or in seawater culture, it must belong to either (1) the Olpidiaceae, (2) the Synchytriaceae, (3) the Plasmodiophorales, or an as yet un-named low group coordinate with the three named. The development of sori in culture strongly indicates a relation to the Synchytriaceae, but it is barred from that group by lack of a uniflagellate zoospore. It is also obvious that *D. marinum* may be related to the Plasmodiophorales, from which it is also barred by lack of the biflagellate zoospore. There is thus no choice but to refer the genus *Dermocystidium* to Rhinosporidiaceae Ciferri, 1932. Ciferri called attention to the close relationship of *Rhinosporidium* and *Dermocystidium* but placed

the latter in the Olpidiaceae under the mistaken impression that its life cycle included a uniflagellate zoospore. *Dermosporidium* should then be placed in the same family, which would be co-ordinate with the Olpidiaceae and Synchytriaceae in the Chytridiales. How many more of the "orphan" genera should be included is problematical but many may qualify.

If *Dermocystidium* is later shown to have a flagellated zoospore, the above placement would be unnecessary, and the genus would be included in Synchytriaceae as suggested by Mackin and Boswell, 1956.

Distribution of *D. marinum*

Distribution in Louisiana. Several hundred samples of oysters, from more than one hundred bays and bayous in the oyster-producing area of Louisiana, have been checked for infection by *D. marinum.* See appendix. Generally the distribution of *D. marinum* in Louisiana is almost the same as the distribution of the host oyster, with a few areas of continual low salinity excepted. For further discussion of salinity effects see the section on epizootiology. Incidence in these samples has varied from zero to 100 percent, depending on the location of the sample, ecological conditions, seasons, and age of the oysters. The stations where zero incidence has occurred are all in areas marginal for oyster production because of mean salinity below 9 ppt or because of seasonal flushing with fresh water or by flooding of the Mississippi River. In Atchafalaya Bay, subject to continual low salinity, *D. marinum* has never been found. In areas such as Sandy Point Bay, during periods of high salinity the incidence and weighted incidence may be high, while in periods of Mississppi River flooding the disease may disappear completely. Areas such as lower Barataria Bay are never free of disease, even in comparatively low salinity periods in winter. Some low incidences recorded in areas where the salinity is above 20 ppt were due to sampling of immature oysters in new plantings.

Data on distribution in Louisiana have been condensed as far as possible and are contained in an Appendix. Exact locations are not given in this appendix, but only the names of bays or bayous, and sometimes of more restricted areas within bays. An explanation of the appendix appears at its head.

Range of D. marinum. The southernmost limit of distribution of *D. marinum* is not known. Of seven live oysters from near Tampico, Mexico, presented to the author by Mr. Albert Collier, five were found to be infected. This is the most southerly record now known. These oysters were collected in January of 1950, and sectioned by the author. It is quite probable that the disease extends beyond Tampico.

Oysters collected by Dr. S. H. Hopkins in 1951 from Fulton Beach, Aransas Bay, Texas, were found to be infected. Several lots of oysters from the same area were found to be negative. Ray (1954b) failed also to find *D. marinum* in that area, but found it in several samples from various parts of Galveston Bay.

Hofstetter and Heffernan (1959) surveyed several Texas bays for *Dermocystidium marinum* infections. Aransas Bay and Galveston Bay were shown to have endemic areas of infection. The fungus was found on five out of six reefs studied in Galveston Bay, and on four out of seven in Aransas Bay. Some of the samples from Aransas Bay showed very heavy intensity of disease at times (Long reef, Jay Bird reef). These records indicate that the fungus may be on the increase in the Aransas Bay area. A few samples

taken in Copano Bay, Lower Laguna Madre, and Matagorda Bay were negative for the fungus.

Several samples of gapers from Mississippi, near Biloxi, have all shown heavy *D. marinum* infections. Ray (1945b) also reported a 75 percent infection rate in oysters from the Biloxi area. Several samples from Mobile Bay have shown very heavy weighted incidence in several reefs in the lower western side of the bay. Ray (1954b) reported that *D. marinum* is common in the area around Pensacola, Florida, and Ingle and Dawson (1953) reported it from the Cedar Key area. Dawson (1955) found that oysters from numerous stations in Apalachicola Bay, Florida, had intensities of infection comparable to those found in lower Barataria Bay, Louisiana.

From Cedar Key, Florida, south along the Florida peninsula, there is no information on *D. marinum* although a few oysters are produced on the west coast as far south as Lee County. Similarly there are no records for the Atlantic coast side, nor are there any for the coast of Georgia. There are several records for the South Carolina coast from the Bears Bluff Laboratories. Oysters from the intertidal zones of Wadmalaw Island, South Carolina, seem to be largely free of *D. marinum*, but oysters in an experimental pond were found by the author to be heavily infected. *D. marinum* is common in North Carolina at Beaufort. The Virginia Institute of Marine Science has extensive records of *D. marinum* in the lower Chesapeake and its estuaries (Andrews and Hewatt, 1957). Most of the James River estuary seed beds are free of infections, probably because of the constant flushing movement of the water. The seaside bays in Virginia appear to be free of infection. Oysters in the Virginia seaside are intertidal and are subject to very low temperatures throughout much of the winter, which probably accounts for the absence of *D. marinum*.

Dunnington (1956) reported that *D. marinum* was found on both sides of the Maryland Chesapeake from about the Patuxent River southward to the Virginia line. Anonymous (1958) report referred to records of *D. marinum* in Delaware Bay dating to 1953. In Delaware Bay the parasite was somewhat spotty in distribution, the infected areas mostly centering around heavy plantings of Virginia seed.

So far as now known, the Delaware Bay area is the northernmost limit of distribution of *D. marinum*. Severe winters seem to be the limiting factor in defining the northward range. However, little effort has been made to determine whether or not *D. marinum* extends to more northerly areas.

Effect of *D. marinum* on the Host Oyster, Experimental Infection

Histopathology of infection. Fungus disease of oysters was first noted because in many cases the oyster meats shrank excessively with heavy infections. Such shrinkage was not confined to gapers, nor were all oysters heavily infected with *D. marinum* shrunken in appearance. Those oysters which had well developed gonads just prior to spawning usually showed no obvious shrinkage. Oysters dying in late summer usually were more or less shrunken, and a slightly yellowish cast to the meats was usual. Fig. 10 shows an excessively shrunken oyster, heavily infected with the fungus. Maceration of the adductor muscle, as shown in this specimen, is sometimes apparent on gross examination, and pus pockets of leucocytic cells are often seen also. Extremely "glassy" or transparent oysters usually are not infected by *D. marinum*. These oysters are diseased but the cause of "glassy" oysters has not been determined.

FIG. 10. An oyster recently dead of *D. marinum* infection. Note the shrunken meat.

Mackin (1951b) and Mackin and Wray (1952) studied the effect of heavy infections of *D. marinum* on the tissues of the host oyster, using oysters in advanced disease for making sections for comparison with sections of normal oysters. The results are summarized as follows:

(1) Light infections have little effect on the host oyster. *D. marinum* cells in lightly infected oysters usually are hypnospores, and few reproductive stages are found. Light infections may be carried for extended periods without proliferation, especially in winter. Light infections usually are found in the gut epithelium.

(2) Heavy infections cause gross destruction of tissues of the host. All types of tissue are invaded. The Leidig cell tissue (Fig. 4A) often contains masses of parasites and may be destroyed over considerable parts of the body. Arterial walls and heart are also susceptible to heavy invasion (Fig. 11A). Destruction of gut epithelium (Fig. 11B) and adductor muscle (Fig. 11C) is commonly found. Ganglia and nerves may be attacked. Abscessed areas of the mantle and palp are characteristic of heavy infections (Fig. 11D). Renal epithelium appears to be the least affected tissue.

(3) Abnormal deposits of ceroid (see discussion below) which causes the yellowish cast to oysters in advanced disease is characteristic of acute infections.

(4) There is no indication that *D. marinum* cells produce exotoxins in any significant amount. The appearance of sections indicates that damage to the host is due almost entirely to lysis of tissues. In some advanced cases, there may be more cells of the parasite than of the host.

Hewatt (1952) noted that infection with *D. marinum* arrested shell growth or caused loss of shell weight from one to eight or ten weeks before death of the oyster from fungus disease. He made weekly weighings of individual oysters under water over a six-month period. Hewatt was able to predict deaths from the diminished growth or loss in shell

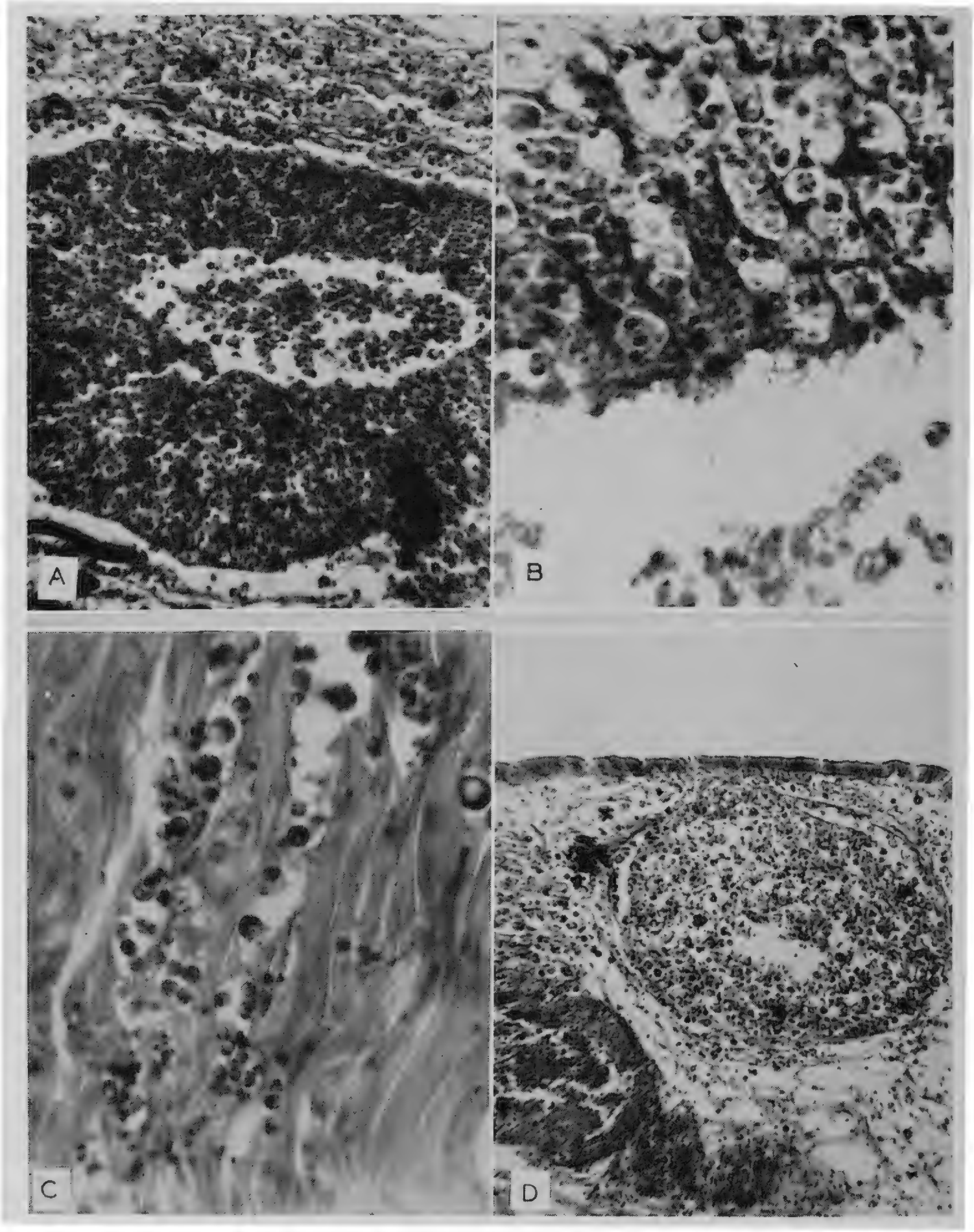

FIG. 11. Sections through tissues of diseased oyster. A. Mantle artery showing masses of *D. marinum* cells in the lumen and surrounding vessel wall which has greatly hypertrophied. B. Numerous *D. marinum* cells in the gut epithelium. Lumen of the gut is at the bottom of the photograph. C. *D. marinum* in the adductor muscle. D. Abscessed area under mantle epithelium.

weight. He observed also that sick oysters failed to open the shell and feed in most cases, and assumed that the closed oysters resorbed shell material in advanced stages of disease.

Menzel and Hopkins (1955a, 1955b) studied the relation of shell growth and increase in weight of oysters to parasitization by *D. marinum*. Shell growth and weight of

individually marked oysters were measured over a two-year period at Bay Ste. Elaine, Louisiana. *D. marinum* infection first slowed growth, then caused it to cease altogether. Their data indicate that the effect of fungus infection may begin eight to eleven months prior to death from fungus disease. Infections acquired in one summer failed to produce death of the host until the following summer.

Ray, Mackin, and Boswell (1953) studied the effect of heavy infections in excised bits of gill tissue. Small pieces of living, heavily infected gill tissue were excised and placed in sterile seawater in Carrel flasks. Antibiotics were used to control bacterial contamination. Control (uninfected) tissues were observed simultaneously. Photographic records were made over the period necessary for destruction of the gill tissues by the fungus, which was about two weeks. This study has been repeated several times. Destruction of the host tissue occurred in all cases.

In the same report (Ray, Mackin, and Boswell, 1953; Reid, 1953), the effect of fungus infections of different intensity on meat weight of oysters, as compared with weights of uninfected oysters, was studied. The normal relation between meat weights of oysters and shell capacity was established for different seasons. Moderate and heavy infections with fungus disease caused significant decrease in meat weight, independent of season. Light infections did not produce significant decreases in meat weights.

Stein and Mackin (1957a) used histochemical methods to determine the effect of *D. marinum* infection on glycogen content of oysters. Sections were made of oysters in advanced stages of disease and compared with similar sections of uninfected or lightly infected oysters. The periodic acid-Schiff (PAS) technique was used for staining as described by Lillie (1954). Malt diastase was used as a test for specificity, since PAS will stain substances in oyster tissue other than glycogen. This study showed that glycogen depletion occurred in heavily infected oysters, but that the effect did not arise from direct selective utilization of glycogen by the parasite. The effect was due to destruction of Leidig cell tissue in which is stored most of the glycogen, and replacement of this tissue by fibrous tissue and masses of leucocytes.

Stein and Mackin (1955) also studied the production of ceroid (lipofuscin, chromolipoid) in diseased oysters. It had been noted (Mackin, 1951b) that the so-called "pigment cells" appeared to increase in oysters in advanced stages of fungus disease. He supposed that the pigment cells contained hemosiderinoid.

To test the hypothesis that increase in the ceroid was associated with heavy infections of *D. marinum* in oysters, counts were made of pigment cells in randomly selected tissue areas around the gut in stained sections of heavily infected, lightly infected, and uninfected oysters. Counts were made in ten oysters of each class and in ten randomly selected fields in each oyster. The mean number per field in heavily infected oysters was 14.59, and in the lightly infected and negative oysters it was 8.30, a statistically significant difference. The presence of excess ceroid indicates an imbalance in fat metabolism, possibly due to an increase in oxidative and reductive metabolic processes as a result of disease.

Ceroid is known to develop in unusual amounts in certain diseases, in some manner connected with defensive host reactions. *Macoma balthica* appears to have a high degree of resistance to infection. Cells of *Dermocystidium sp.* in that host degenerate in cyst-like cavities amid increasing amounts of ceroid. Much of the ceroid seen in sections of oyster tissue may result from degeneration of dead *D. marinum* cells.

Weighted Incidence of Fungus Disease in Gapers and in Live Oysters. Studies compar-

ing intensity of disease in dying oysters with the intensity of disease in survivors have been made at several field stations. Where lysis of tissue rather than exotoxin is responsible for the main damage to the host, it may be assumed that oysters dying have measurably heavier infections than survivors.

A numerical system for estimating the intensity of infections in oysters has been widely used (Mackin, 1952a; Ray, 1954a, 1954b; Andrews and Hewatt, 1957). At present the different numerical grades of infection are defined as follows:

5. A *heavy* infection is one in which the fungus cells are concentrated in great numbers in all tissues. Thioglycollate-cultured tissues appear macroscopically greenish-blue to black in color when iodine solution is applied.

4. *Moderate to heavy* infections are those that appear to be intermediate between the moderately heavy infections and those that are obviously heavy.

3. A *moderate* infection is one in which the tissues are heavily infiltrated in some areas, but lightly dispersed in others.

2. A *light to moderate* infection is intermediate between a light infection and a moderate infection.

1. A *light* infection is one in which the fungus cells number between 10 and 100 in a preparation. Generally speaking small local concentrations of numerous cells are not considered as light infections.

0.5 When fungus cells in one preparation number less than 10 in an entire preparation it is classified as *very light*. Usually a search is necessary to find the few fungus cells present.

0. *No infection.*

Many studies suggest that some oysters probably die of *D. marinum* infection when the infection intensity is only moderate. Because moderate infections are those in which there are local areas of heavy infection, it is assumed that chance, early, acute attacks on vulnerable organs cause these deaths.

Under the weighted system described above, 5.00 is the highest possible score, occurring only rarely where all oysters of a sample are heavily infected. A population of live oysters with a weighted incidence of 2.00 contains an intense epidemic, and more than half of the population may be in advanced stages of disease, with all of the individuals infected. Samples of gapers usually score between 3.00 and 5.00 while the majority of random samples of live oysters in Louisiana scored from 0.50 to 1.50. (See appendix for exceptions.)

TABLE 2

Comparison of weighted incidence and incidence of infection by *D. marinum* in live oysters and gapers at the Bayou Rigaud Station, Louisiana

Study no.		Gapers or live oysters	No. oysters	Incidence of *D. marinum* percent	Weighted incidence of *D. marinum*
1	March to July, 1950	Gapers	46	100.0	4.72
		Live	45	30.0	0.60
2	Jan. to Sept., 1951	Gapers	113	86.7	3.99
		Live	199	48.7	0.88
3	March, 1952 to May, 1953	Gapers	298	95.6	4.37
		Live	478	72.2	1.36
4	Feb., 1955 to Oct., 1956	Gapers	59	92.3	4.28
		Live	106	57.5	0.81

In Table 2 are compared weighted intensities of gapers with weighted intensities of survivors in the populations at the Bayou Rigaud Station in four different periods from 1950 to 1956. All oysters were kept in trays on racks at the laboratory in Bayou Rigaud. Averaging all four studies, the value for the gapers is more than four times the value for the live oysters. There is no overlap, the highest value recorded for survivors (in the 1952–53 study) had a weighted intensity of 1.36 while the lowest value for gapers (in 1951) was 3.99.

A further field study of intensity of infection in live and dead oysters was made in 1957. Seven field stations were set up in Billet Bay, Lake Grande Ecaille, and Barataria Bay. At each station 1000 oysters were placed in March, 1957, and gapers were collected weekly. One thousand eighty-eight gapers were collected from these seven stations in the period from March to September inclusive. These gapers had a weighted incidence of 3.64 and an incidence of 92 percent infection with fungus disease. Eighty samples of live oysters, usually 25 oysters to a sample, were collected from the seven stations and checked for disease at approximately monthly intervals. A total of 1972 live oysters were checked for incidence and weighted incidence of fungus disease. The incidence was 70.0 percent and the weighted incidence was 1.08 (Table 3).

TABLE 3

Weighted incidence in gapers and live oysters at seven stations in Lake Grande Ecaille area and Barataria Bay, Louisiana, March 1957 to Sept., 1957

Gapers or live oysters	No. oysters	Incidence of *D. marinum* percent	Weighted incidence of *D. marinum*
Gapers	1088	85.0	3.64
Survivors (80 samples)	1972	70.0	1.08

Andrews and Hewatt (1957) found similar conditions to exist in the York River. Data from their study are presented in Table 4. The low-weighted incidences are from winter periods, when *D. marinum* nearly disappears from the oysters in Virginia waters. The high weighted incidence of *D. marinum* in gapers indicates that nearly all deaths from disease in the Virginia Chesapeake area were from fungus disease. Predators were largely eliminated from their suspended trays.

TABLE 4

Weighted incidence of fungus disease in live oysters and gapers in the Chesapeake Bay area April, 1953 to December, 1955

Data from Andrews and Hewatt, 1957

Gapers or survivors	No. oysters checked	Percent infected	Weighted incidence
Gapers	2634	92.0	4.27
Survivors (130 samples)	3857	34.0	0.53

Weighted Incidence of Fungus Disease and Death Rates of Oysters in Experimental Tray Populations. A measure of pathogenicity of fungus disease is also provided by the weighted incidence—death rate correlation. Since weighted incidence normally increases with rising temperature from early spring to late summer, an opportunity is afforded to relate death rates and intensity of disease in oyster population.

Andrews and Hewatt (1957) published extensive data on weighted incidences and death rates of oysters held in trays in the York River. One of the oyster populations was taken from the James River seed area and the oysters were uninfected when placed in trays at Gloucester Point in the York River. A moderate mortality occurred in these oysters in their first summer; in the second summer of exposure to disease, mortality rates were excessive. The data from Andrews and Hewatt's paper were extracted and put in graph form in Fig. 12. The correlation between death rates and weighted incidence of disease is obvious from the graph.

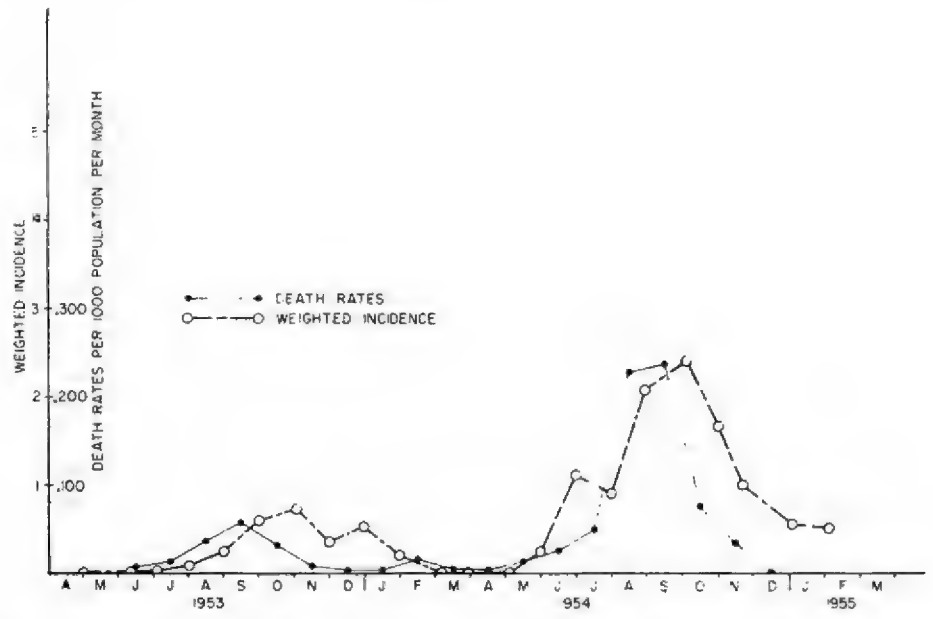

FIG. 12. Death rates and weighted incidence of infection of oysters with *D. marinum* in the York River, 1953 to 1955. Data from Andrews and Hewatt (1957). Oysters were uninfected when placed in trays in the spring of 1953.

Similar studies have been carried out in the Louisiana area (Mackin and Sparks, 1961). The graph, Fig. 13, shows weighted incidences and death rates at three selected stations, one in Billet Bay, one in Lake Grande Ecaille, and one in Barataria Bay. The study began in March and ended in September, 1957. One thousand oysters were placed in trays at each station and both the weighted incidence of disease and the mortality rates were recorded at regular intervals for this period. The data in Fig. 13 are plotted

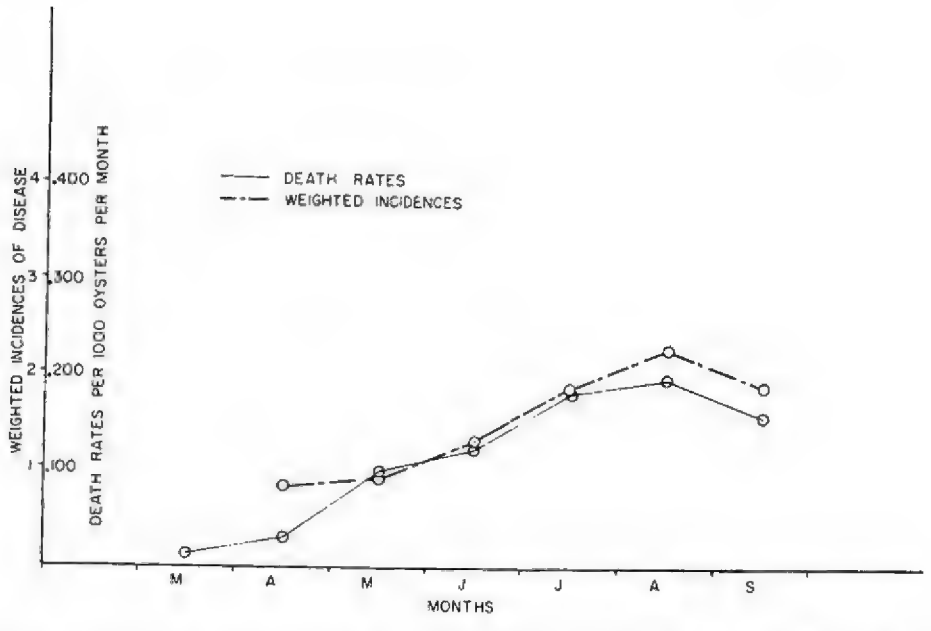

FIG. 13. Weighted incidences and death rates at three stations (Billet Bay, Lake Grande Ecaille, and Barataria Bay). Data are averaged for the three stations. From Mackin and Sparks, 1959.

on a monthly basis as a means of smoothing the curve, although mortality data were taken weekly. Death rates and weighted incidences for the three stations are averaged. The fidelity with which death rates follow the weighted incidences is remarkable. This type of correlation has been noted for a number of stations in Louisiana.

Field Records of Disease Correlated With Mortality Rates. Early in the field studies it was found that oysters held in trays at certain stations consistently had low mortality rates and that others held at other stations had consistently high mortality rates. It was reasoned that if *D. marinum* was a predominant cause of mortality, it should be possible to detect significant differences in intensity of infection between tray populations with contrasting mortalities. A study was made in 1948–49 to test the hypothesis. Oysters from the Chene Fleur Bay, a low mortality area, were used to stock five stations, including Chene Fleur, with 1000 oysters each. Mortalities were checked over a 14-month period, and all gapers possible were collected and sectioned to determine the incidence of disease and intensity of infection (Mackin and Wray, 1952). At the end of the study a sample of live oysters was also sectioned and the incidence of disease established. The relation of incidence and intensity of disease to mortality rates is shown in Table 5. At

TABLE 5

Relation of incidence and intensity of fungus disease in oyster populations, and death rates in those populations. Approximately 1000 oysters were placed at each station and all were stocked with oysters from Chene Fleur Bay

Station	Death rates per 1000 per 14 months	Gapers recovered	Gapers in advanced disease percent	Live oysters sampled for disease	Incidence of disease in live oysters percent	Weighted incidence in live oysters
Chene Fleur (Control)	0.297	3	33.3	26	0.0	0.00
Bassa Bassa	0.614	17	64.7	21	38.1	1.05
Grande Ecaille	0.766	13	100.0	21	52.4	0.71
Sugar House	0.825	15	100.0	23	52.1	0.87
Bayou Rigaud	0.842	21	90.5	21	66.7	1.14

the low mortality station, no disease was found in live oysters, but one gaper was heavily diseased, showing that *D. marinum* was present but relatively rare. Although all stations were stocked with oysters from Chene Fleur Station, at stations where high death rates were usual the oysters developed proportionately higher incidence and intensity of disease.

Owen (1955) used the same general method. In his study he set up a series of stations in high mortality areas and contrasted these with a control, a low mortality station in Sister Lake. Owen's study ran for seven months and mortalities were severe. He placed two types of oysters at each station. Half were from high fungus-incidence areas (Barataria Bay and Pensacola Bay, Florida), and half were from low incidence areas (Sister Lake and California Bay). His data in Table 6 have been condensed from several tables to be comparable to the data in Table 5. Owen found the same correlation of high incidence and high mortality as was found at our own stations. Some of Owen's stations were in the same area as our own (Sugar House and Grande Ecaille).

Measurement of Pathogenicity of D. marinum Infection, Using Death as a Criterion (Tables 7–13). Experiments to determine if *D. marinum* infection could be lethal to oysters began very early in the studies. Prokop (1950), working at the Grand Isle Laboratory, described four experiments in which oysters were subjected to infection by

TABLE 6

Comparison of intensity and incidence of disease in oysters at a low mortality station (Sister Lake) and in oysters in high mortality areas from the Mississippi River to Bayou Lafourche
Data condensed from Tables 50 to 53 of Owen (1955)

Station	Death rate per 1000 in 7 months	Gapers recovered	Gapers in advanced disease percent	No. live oysters studied	Incidence of fungus disease in live oysters	Weighted incidence in live sample
Sister Lake (Control)	0.290	7	0.0	20	60.0	0.30
Sugar House Bend	0.720	6	100.0	20	100.0	1.85
St. Mary's Point	0.400	6	66.7	20	75.0	0.87
Grande Ecaille	0.720	10	100.0	20	100.0	1.90
Adams Bay	0.610	12	100.0	20	85.0	1.22
Bayou Scofield	0.580	4	100.0	20	100.0	1.50
Sandy Point Bay	0.580	6	100.0	20	90.0	1.45

D. marinum. In the first of these, purulent material from an infected oyster was injected into the adductor muscle of experimental oysters and sterilized material into controls. Eighty-five percent of the experimental oysters died and 50 percent of the controls in four months. In the second study, experimental oysters were merely placed in an aquarium where diseased oysters had been held, and the controls placed in a sterilized aquarium. In this one, more oysters died in the control than in the experimental group. In the third experiment, oysters were inoculated with purulent material from fungus-infected oysters; mortalities were 100 percent in experimental aquaria in 25 days and 50 percent in controls. In another repetition 66.6 percent of the experimental oysters died and 100 percent of the controls.

In these studies initial infection varied between experimental and control oysters used. Also, all experiments were in aquaria with running seawater containing infective cells of *D. marinum*. As a result oysters died in large numbers in both experimental and control aquaria. In addition, infection methods were poor in that infective cells were injected into the oyster tissues. It is now known that injection is not as effective as feeding. Nevertheless, infection was accomplished in two of Prokop's studies. The significance of differences between controls and injected animals was uncertain.

By 1950, however, understanding of the factors involved in infection had increased, and the first successful laboratory experiment involving infection and induced mortality was carried out (Mackin, 1952b).

In the period July 31, 1950, to October 16, 1950, a study was carried out at the Grand Isle Laboratory which demonstrated (1) that the infective elements of *D. marinum* infection are water-borne, (2) that infection may be accomplished by placing diseased oysters (carriers) in proximity to non-diseased susceptibles, and (3) that a high percentage of oysters thus infected die.

Seawater at the laboratory was passed through an overhead constant-head tank, which also served as a settling tank for silt, and into two aquaria below. The experimental tank contained 20 oysters from a group with known high incidence of infection with *Dermocystidium*. A sample sectioned prior to the study was 86 percent infected, and the weighted incidence was 2.07. The oysters originally were caught as spat on cemented egg crate fillers in the spring of 1948, stored in trays at the Chene Fleur station until the fall of 1949, and brought to Bayou Rigaud. The oysters were therefore more than two years old at the beginning.

In the control tank were placed 20 oysters, which were known to have a low level of incidence of *Dermocystidium*, established by sampling at less than 5 percent. These oysters were brought directly from the Chene Fleur racks and had never been in the lower bay area before.

From each of these tanks a tube led to a rubber manifold from which tubes distributed water to 16 finger bowls each containing one oyster from the same lot as the oysters with low infection incidence in the control tank. With individual oysters in separate finger bowls, the possibility of their infecting each other was excluded. Fig. 14 shows part of the control unit, the manifold, and finger bowls with the experimental unit at the right.

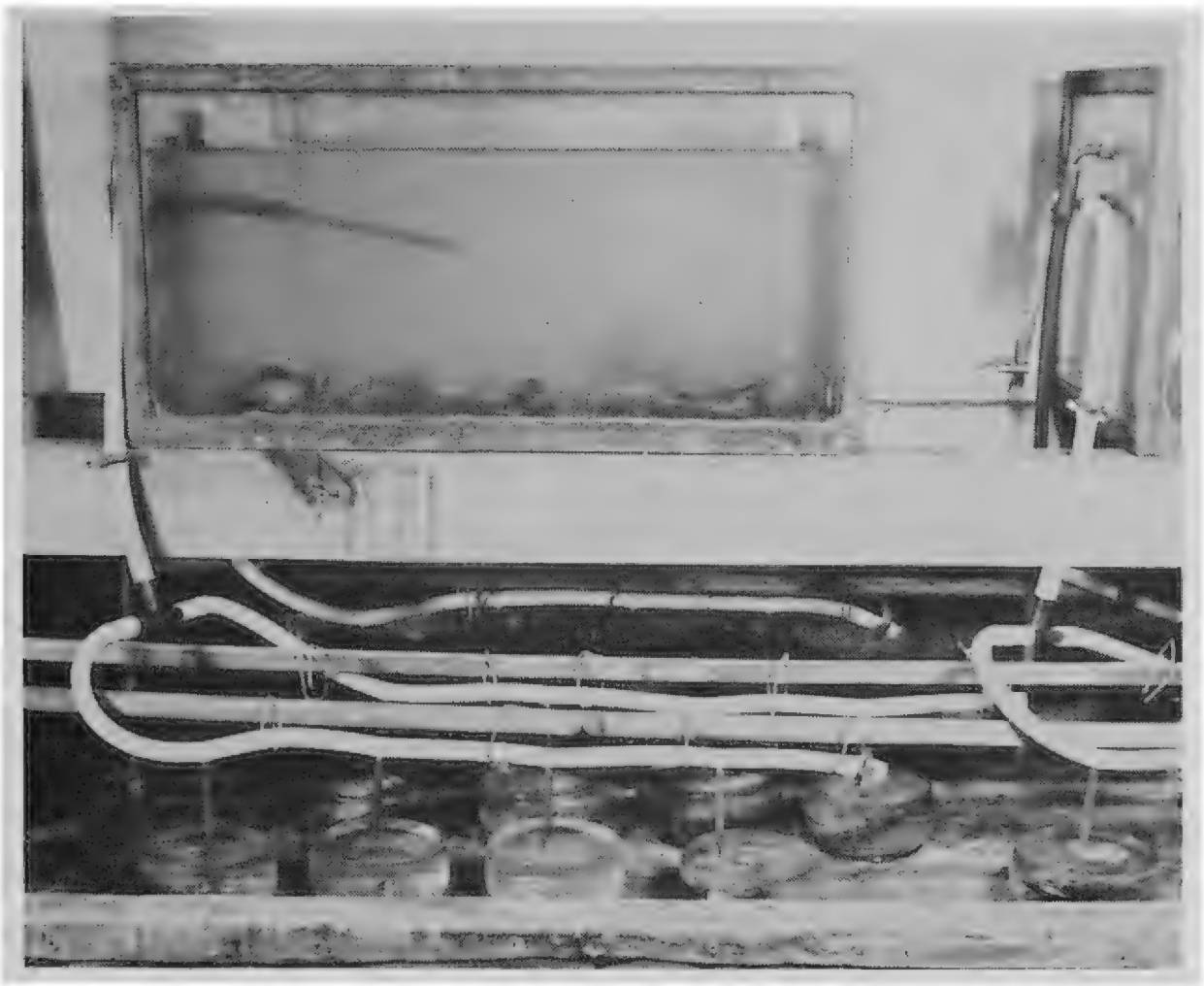

FIG. 14. Detail of experimental apparatus showing method of demonstration that *D. marinum* infective cells are water-borne. Oysters in the upper tank (end showing at right) were heavily infected and passed the fungus to the oysters in finger bowls below. The unit shown is the control, which was identical in set-up to the experimental tank (except for the percentage and intensity of infection of the oysters).

As completed the two units were receiving water from a common source. In the experimental unit, water passed through a tank containing oysters known to have high incidence of *D. marinum* and was delivered to 16 low-level-incidence oysters in individual finger bowls. The other control unit was precisely the same except the intermediate tank contained oysters with very low incidence of infection. The experiment was carried on in enzootic territory where the water necessarily contained infective elements, so that increasing infection of control oysters as the study progressed was considered unavoidable. However, it was reasoned that placing infected oysters in the flow stream of the experimental group should produce greater incidence of disease than in the controls, provided the infective element was water-borne and that the aquarium contained all necessary elements for completion of the life cycle.

TABLE 7

A study of transmission of infection of oysters by *D. marinum*
July 31, 1950 to October 16, 1950

Aquarium	Type	No. oysters	Not recovered for sectioning	Weighted incidence in those recovered (live and dead)	Percent mortality
20 oysters with high percentage of carriers in experimental tank	Gapers	15	2	3.83	75.0
	Survivors	5	0		
16 susceptibles receiving water from the carrier tank in experimental finger bowls	Gapers	8	1	2.40	50.0
	Survivors	8	0		
20 oysters with low percentage of carriers in the control tank	Gapers	3	0	1.15	15.0
	Survivors	17	0		
16 susceptibles with low infection incidence in the control finger bowls	Gapers	4	0	0.69	25.0
	Survivors	12	0		

Results of this study are presented in Table 7. Intensity of infection in the gapers and survivors is given. The carrier oysters in the large tank, known to have a high incidence of infection with *Dermocystidium*, had by far the heaviest mortality (75 percent), and the oysters in the finger bowls fed water from the aquarium containing this group were next (50 percent). These figures may be compared with mortality of 15 percent in the large control aquarium and 25 percent in the control finger bowls. The weighted incidence of the experimental oysters in finger bowls was 3.48 times greater than that of the control oysters in finger bowls. This difference was statistically significant.

The weighted incidence of disease in all gapers from both experimental and control aquaria was 4.26, and the weighted incidence of all survivors was 0.57. These frequencies were similar to those in the field studies. Two of the 16 control oysters developed advanced disease and died.

It was recognized that experimentation would be improved if a supply of completely disease-free oysters could be found. The early source of oysters with low level of infection was Bay Chene Fleur, but field data showed the level of incidence in those oysters to be increasing. It became necessary to procure experimental oysters from a more northerly area on the east coast of the United States outside the range of *D. marinum* and from a local area where ecological conditions were unfavorable to *D. marinum*.

Ray (1954a, 1954b) demonstrated that *D. marinum* could be successfully infected in closed aquaria by a "proximity" method, i.e., by placing infected oysters in an aquarium with uninfected oysters, and also by a "feeding" method in which meats of oysters dying of *D. marinum* infection were mixed with water in a closed, aerated aquarium containing uninfected oysters. It was later demonstrated at the Grand Isle Laboratory that water for *closed*-aquarium experiments could be taken from endemic areas with little danger of infection to experimental oysters due to the small number of infective cells in the volume of water needed to fill a small aquarium. Ray obtained disease-free oysters from the Milford, Connecticut Laboratory of the U.S. Fish and Wildlife Service. Data from experiments using these methods are presented in Table 8. Mortality was greater in experimentally infected oysters.

Repetitive studies using the feeding method were carried out at Grand Isle Laboratory (Ray and Mackin, 1954; Ray, 1954a; Ray and Chandler, 1955). In one, oysters used

TABLE 8

Percent of oysters dead, surviving, and infected from experiments in closed aerated aquaria (Ray, 1954a). Uninfected oysters used in these experiments were obtained from Milford, Connecticut. Oysters used for infecting experimental oysters were obtained from the Grande Isle, Louisiana, Laboratory and Galveston Bay, Texas. Studies ran for 21 weeks

	Experimental oysters				Control oysters			
	No.	Dead	Survivors	Infected	No.	Dead	Survivors	Infected
Infection by proximity	40	50	50	95	15	20	80	0
Infection by feeding	30	83	17	87	15	40	60	0

were from Connecticut; in the others oysters came from Redfish Bay, Louisiana, which at that time was free of disease. The data from these repetitive experiments are condensed in Table 9. Mortality was greater in the experimentally infected oysters. The

TABLE 9

Repetitive studies of effect of infection of oysters with *D. marinum* using the "feeding" method

Study no. Origin of experimental and control oysters Period of the study (1953)	1 Redfish Bay, Louisiana July 16 to Sept. 11	2 Redfish Bay, Louisiana July 16 to Sept. 18	3 Milford, Connecticut June 11 to July 16
No. oysters in experimental tanks	30	25	25
No. dying in experimental tanks	25	23	24
Percent dying in experimental tanks	83.3	92.0	96.0
Weighted incidence of all experimental oysters, including survivors	4.34	4.40	3.40
No. oysters in control tanks	60	25	25
No. dying in control tanks	6	6	13
Percent dead in control tanks	10.0	24.0	52.0
Weighted incidence in control tanks	0.0	0.0	0.0

experiment using oysters shipped from Connecticut was faulty because of a large number of deaths in control oysters due to weakened condition. However, controls remained free of infection.

This study (Table 9) differed in that running filtered seawater was used rather than closed, aerated aquaria. The filtration apparatus to prevent infection of control oysters in aquaria supplied with running seawater, in endemic areas, had been perfected in the spring of 1953 at the Grand Isle Laboratory. In its final form it consisted of (1) a settling tank, and (2) three tandem filters made of crisscrossed glass wool fibers packed in glass containers. There were three sets of these in use simultaneously (Fig. 15). The water stream from the last filter of a set was divided between a control tank and an experimental tank.

On March 26, 1953, two groups of oysters of 15 each were "injected" with tissue mince of heavily infected oysters (each group with mince from a different oyster). Tissue mince was diluted with seawater (100 cc seawater to 1 cc tissue mince), and about 1 cc of the dilution was injected with a pipette or small syringe into the shell cavity of the experimental oysters. No effort was made to find uninfected populations for this study. Infections were made on previously infected populations, using short periods for incubation of the parasites in the oysters. A control group of 26 oysters was drawn from the same population, without injection. This first experiment was actually a test of the "injection" method rather than of the efficiency of the filters, since the

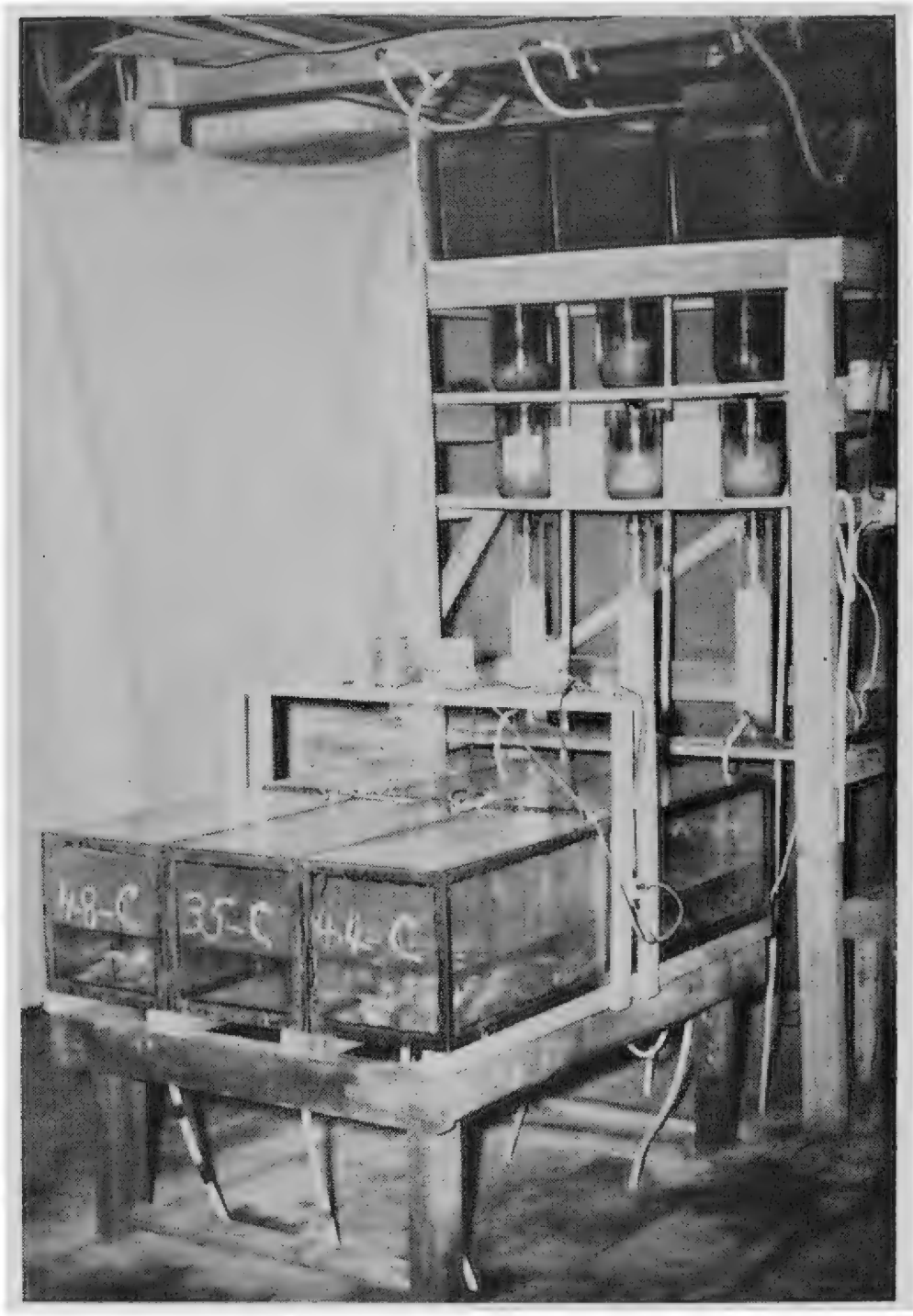

FIG. 15. Experimental equipment for testing for pathogenicity of *D. marinum*. Settling and constant head tanks are at the top of the picture; below these are three filters for each experimental unit. Water from the bottom filter was divided between the control tanks (foreground) and experimental tanks. Three complete experimental units are shown.

control population was already infected. The filters merely prevented additional infection of controls (and experimental oysters). The results are presented in Table 10.

TABLE 10

Study of the effect of injecting tissue mince from oysters with heavy infections of *D. marinum* into the shell cavity of experimental oysters. Study began March 26, 1953, ended 12 days later on April 10, 1953

	No. oysters used	Weighted incidence after 12 days
Experiment 1, injected	15	2.07
Experiment 2, injected	15	2.56
Control oysters	26	1.65

Injection did increase the weighted incidence of the experimental oysters. In the short period of the study only one oyster died (in Experiment 2), and the increase in weighted

incidence was used to test the efficiency of the infection method. No data were developed on the efficiency of the filters but test plating for bacteria in the seawater before and after passage through the filters showed a decrease of about 99 percent in passage.

Using the filtration method and the injection method, three experiments were set up in three sets of aquaria receiving filtered seawater. Oysters used in these three studies were all disease-free at the beginning of the studies. In two studies disease-free oysters from Milford, Connecticut, were used; in the other, oysters from Redfish Bay, Louisiana, were used. The data derived from these three studies are presented in Table 11.

TABLE 11

Summary of the results of three experiments (30, 35 and 48) in which infection was accomplished by using the "injection" method and infection of control and experimental oysters was prevented by use of glass wool filters set in the seawater stream. All oysters were disease-free as shown by tests of samples of the experimental populations prior to the study (65, 70 and 98 oysters respectively). Experiments in the period from June 10 to September 18, 1953

Dates	Experimental animals				Control animals		
	No. oysters used	No. dead	Dead percent	Case death ratio	No. used	No. dead	Dead percent
June 10 to July 16, 1953	25	24	96	0.960	24	8	33
July 16 to August 25, 1953	24	23	96	0.960	23	6	26
August 13 to Sept. 18, 1953	30	28	93	0.932	30	2	7

In these experiments only one control oyster contracted disease. It is not known whether this one was infected through the filters or in some other manner. In any case, the studies showed a high rate of death in diseased oysters, so high that there can be no mistaking the extreme pathogenicity of the fungus under conditions of the experiment. All oysters used in these studies were probably susceptible and could not have had the resistance or immunity expected in oysters from endemic waters where natural selection might have eliminated susceptible stock. The fungus was proved to be very highly pathogenic. The field studies (in trays) and observations of field mortalities on planted beds indicated that the difference in aquarium oysters and those in natural habitats is a matter of time, death taking place under aquarium conditions in relatively less time.

The infection method used in the 1953 experiments proved to be satisfactory. The data show that at least 97 percent of the experimental oysters were infected and that they probably were infected within one day. This opened up new fields of investigation. It was proposed that the rate of development of disease in oysters was a function of the number of infective cells received in a "dose". Studies were set up in which different dosages of infective cells were given to experimental oysters of a number of groups. Two groups of experiments were carried out, testing different ranges of dosage. Counts of cells of *D. marinum* in diluted tissue minces made up from heavily infected gapers were made. A standard haemocytometer was used for the counts. Aliquots of the tissue mince were cultured in thioglycollate medium for a short period (one day) to enlarge the cells for recognition purposes. Ten aliquots were counted, using ten fields for each, and the results were averaged. The tissue minces were then diluted with sterile seawater to produce a stock suspension of 5×10^6 cells per cc. This stock solution was then diluted further to produce the concentration of cells used in the experiments. Parts of this study, those experiments using dosages from 5×10^4 to 5×10^6, were carried out in the

summer of 1954, the remainder in concentrations of 10 to 5000 were carried out in the summer of 1955. One control (oysters injected with tissue mince of uninfected gapers) was carried with each group of five experiments.

TABLE 12

Relation of infective dosage of *D. marinum* cells to death rates of oysters in experimental aquaria. Studies lasted 41 days

No. oysters used in experimentation	No. infective cells injected per cc of tissue mince	Mortality in percent per day	Days to first death to fungus disease
25	0.00 (Control)	0.10	....
18	0.00 (Control)	0.27	....
18	10	0.27	....
18	50	0.27	....
18	100	0.27	37
18	5×10^2	0.80	36
18	5×10^3	0.95	27
25	5×10^4	1.27	17
25	2×10^5	1.95	21
25	5×10^5	2.34	14
25	2.5×10^6	2.17	13
25	5×10^6	2.17	16

The results of these studies are presented in Table 12 and Fig. 16. The data in Table 12 indicate that in the period of the study (41 days) concentrations of infective cells from 10 to 1000 had no significant effect on the oysters.

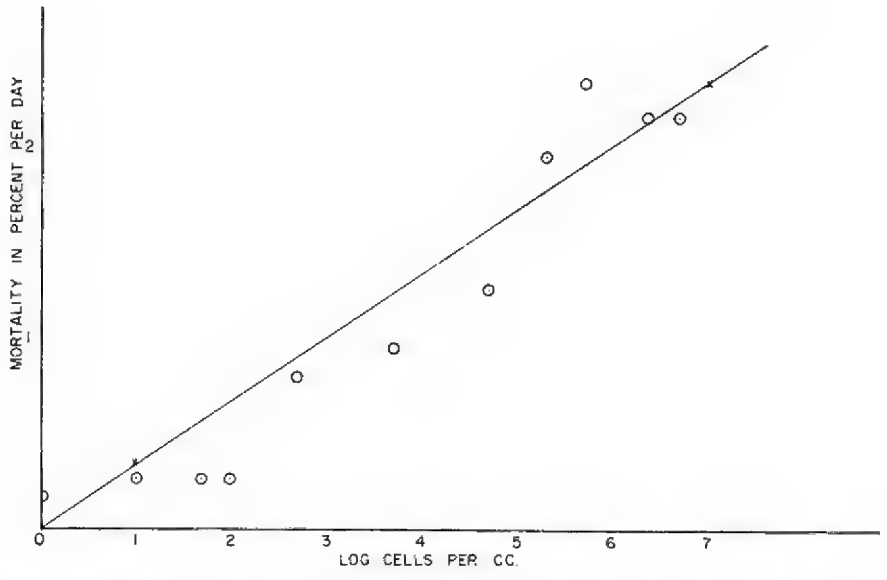

FIG. 16. Increase in death rate as the number of infective cells given in a dose is increased. The data indicate that very low dosage may not increase death rate within the period of the study (41 days), and similarly, that after a certain high dosage is reached, continued increase may have little effect.

The table also shows that the heavy dosage of 5×10^6 produced no significant increase in mortality rate over a dosage of 5×10^5. The time of death of the first heavily infected oyster did not differ materially in these heavy concentrations, nor did the death rate for the entire study vary much. Intermediate dosages, however, showed a regular progressive increase in death rates in relation to dosage. Fig. 16 shows the calculated increase in death rate in relation to dosage. Death rates in percent per day were plotted against the logarithm of the dosages. An increase in death rate of 0.33 percent per day per log unit of the dosage was found. A study of these data indicates a high degree of

pathogenicity under conditions of the experiments. The case-death ratios in the higher dosages (5×10^4 upward) ranged from 0.920 to 1.000.

Andrews and Hewatt (1957) used the injection method to test the effect of *D. marinum* infection on oysters in Virginia. The studies were designed to repeat studies by Mackin and Ray, as reported by Ray (1954b). They used oysters from Deep Water Shoal in the James River which were fungus-free. They used two experimental groups and two control groups of 19 to 22 oysters each. One of the control groups (4) in Table 13 was suspended in the water at Gloucester Point on the York River, where they were subjected to natural infection. The others were held in closed aquaria. The results of these studies by Andrews and Hewatt are presented in Table 13.

TABLE 13

Effect of infection of Virginia oysters by the injection method. After Andrews and Hewatt (1954). Studies were in the period from June 22, 1954 to July 29, 1954

Group no.	No. of oysters used	No. dead in experimental period	Percent dead	Weighted incidence of gapers	Weighted incidence of survivors
1 Experimental	22	15	68	3.80	4.43
2 Control	20	4	20	0.00	0.00
3 Experimental	20	18	90	4.50	5.00
4 Control	19	5	26	5.00	3.57

These data are similar to those developed by experimentation in Louisiana. It is interesting to note that the control group of animals placed in a tray in the York River developed slightly more mortality than did the control group held in an aerated aquarium, the difference being caused by natural infection of the oysters held in the tray.

Physiological Tests of Pathogenicity. Physiological tests of pathogenicity of parasitic organisms have been widely used. These usually measure variation from normal or vital functions of infected hosts. Normal level is established by uninfected controls. Another effective method of testing for pathogenicity is to measure the capacity of infected organisms to withstand detrimental environmental conditions as compared with uninfected controls. Both of these methods have been used in testing the effect of *D. marinum* infection in oysters.

Testing for Weakness of Oysters in Advanced Disease When Held Out of Water. It was hypothesized that oysters in advanced stages of disease caused by *D. marinum* were less resistant to holding out of water than were oysters with less advanced or no disease. Since it was impossible to determine whether or not oysters were in advanced disease without destroying them, it was decided to begin with a group of oysters with known high incidence of disease but in which intensity of infection in individual oysters was unknown. Initial infection level in the population as a whole was determined by sampling and the population then subjected to abnormal conditions. If heavily infected oysters were more susceptible to the treatment than were lightly infected or negative oysters, the population might be expected to develop differences in infection during treatment. Those oysters gaping earliest would show greatest susceptibility to the treatment. It was arbitrarily decided to call the first 25 oysters to gape the "experimental" oysters and to take a random sample of non-gapers from the remaining population for the "controls." When 25 oysters had gaped, tissues from "experimental" and "control" oysters were taken and cultured in thioglycollate by the Ray method in order to determine the

intensity of disease, and the intensities in experimental and control groups were compared. Data from two such experiments are presented in Table 14.

The first study was set up on April 21, 1953. A sample of the experimental population was taken at 0900 and thioglycollate cultures were made to determine pre-experiment disease incidence. Three-year-old oysters from the laboratory racks were used in this study. At the same time, 250 oysters from the same group were spread out on the floor of the laboratory. The first 25 oysters to gape made up the experimental group and the gut tissues of these were cultured as they gaped. The last gapers were collected on April 26, 1953, at 2030 (131½ hours). The control sample was taken from the survivors at that time. These were selected for tight closure of the shell. The period of this study was relatively cool, and the laboratory aquarium room was not heated.

The oysters used in the second experiment were 20 months old and had been raised from spat at the laboratory. Four hundred thirty-five oysters were used not counting the sample taken prior to treatment. The period was May 6 to 9, 1953, and the weather was warm. All procedures were the same as in the first experiment except a close check was kept on the order of gaping. The experimental oysters gaped much more rapidly in the second study than they did in the first, probably because of the higher air temperature and the slightly higher original intensity of disease in the experimental oysters.

The data indicate that the treatment produced acceleration of disease in the oyster populations used. They also show that oysters with heavy infections gape earlier, and have less resistance to holding out of water than do those with lesser infections. In the first study, the pre-experimental sample showed that about one in 25 oysters of the experimental population was heavily infected. There were about 10 heavily infected in the population of 250, and 11 heavily infected oysters gaped when the 250 were held out of water. The control sample taken from the non-gapers indicated that there were no heavily infected oysters remaining in this group.

Ciliary Activity in Diseased and Non-diseased Oysters. Observations of diseased oysters indicated that in late stages of disease, feeding rates were reduced. Sections usually showed that the gut of most heavily infected oysters was empty. Studies by Hewatt (1952) indicated that oysters in advanced stages of fungus disease kept the shell closed most of the time and therefore were probably feeding less than oysters with light disease or no disease. It was decided to test the efficiency of ciliary action of the gills, as an indicator of reduced respiratory and feeding efficiency.

Lund (1949, 1957) compared the ciliary capacity of diseased oysters for production of faecal and pseudofaecal matter with that of normal oysters. He used oysters shipped to him from Barataria Bay by the writer and these oysters probably included some with fungus disease. However, the oysters were mostly young ones and it is probable that

TABLE 14

Comparison of weighted incidence of fungus disease of gapers and survivors when held out of water. Experiments 1 and 2 used different populations of oysters

Exp. no.	Air temp. range, °C	No. oysters in experimental population	Weighted incidence of population		
			Pre-experiment	In gapers from the experiment	In non-gapers (control)
1	15 to 25	250	1.66	3.20	1.72
2	18 to 27	435	1.70	2.69	2.20

Lund was dealing also with some disease in addition to *Dermocystidium*. Only those which were visually diseased in advanced stage by the end of the study were placed in the diseased group. He compared these with another group (controls) which may have included oysters lightly infected with various diseases as well as some which were actually "healthy". Lund's study probably provided the first experimental assessment of a disease on a commercially valuable invertebrate, in terms of physiological derangement. The present study corroborated Lund's finding and by careful diagnosis measured the effect of one specific disease as follows.

The experimental tank devised by Lund (1949, 1957) was used in these studies. By Lund's method, the ciliary activity of control and experimental oysters is measured by means of a lucite trough which collects the faecal and pseudofaecal matter precipitated by a series of oysters. The trough was divided into three smaller troughs, running lengthwise, and each of these three was divided into six compartments in linear series. Each compartment was designed to contain six small oysters. However, oysters as small as those used by Lund have only light infections of fungus disease. It was necessary in this study to use oysters of larger size, and only three of these larger oysters could be placed in each compartment. In one of the three troughs we placed only shells of oysters, in order to measure the amount of silt precipitated without the action of oyster gills and mantles. The control trough and the experimental trough each contained 18 oysters. Each experimental oyster was matched for size by a control, and the two of each matched pair were placed in the same relative position in their respective troughs. Sizes ranged between 2½ and 3½ inches in length.

The experimental oysters for the *first* study were taken on April 9, 1952, from a planted bed at Philo Brice light in Lake Raccourci. A check of a sample of thirty oysters by the thioglycollate method showed that approximately 67 percent of the experimental oysters were infected, and that the weighted incidence was 1.60. The experimental oysters were kept in trays or racks under the laboratory until the beginning of the study on April 26, 1952. They were planted in September of 1951, and thus were probably about 20 months old.

The control oysters were brought from the Chene Fleur station April 25, 1952, and were kept for the intervening two days in a running water aquarium in the laboratory. These oysters were from a group which set in May of 1951, and thus were approximately 12 months old. Although younger, they were about the same size as the experimental oysters. A sample of 25 of these oysters was examined; none were infected. Twenty-five additional oysters were examined a few weeks later and again were all negative.

Oysters and shells were carefully cleaned of all adhering silt before placing them in the trough with running sea water. Water was fed into the three troughs in equal measured amounts from a single constant-head tank, each trough receiving a flow of 1000 cc per minute. The actual amount of turbid materials suspended in the water varied during the ten days of the study, but was the same within narrow limits in all three troughs at any one time. Turbidities in the experimental troughs were often high, but were never as high as they were in Barataria Bay at the same time under natural conditions. The heavier silt particles settled out in the seawater cistern and constant-head tank before reaching the experimental troughs.

On May 5, 1952, the water flow was shut off, and the oysters and shells were removed from the trough after being carefully cleaned in the water in their respective compartments. The pseudofaecal and faecal matter were allowed to settle, were removed from

each compartment, and were transferred to graduates for measurement. The graduates were set aside for 24 hours to allow the materials to settle before reading the amount.

Silt from the trough containing the shells was similarly collected and measured. The amount of settling out in the compartments containing shells was subtracted from the amount in corresponding compartments containing oysters. The difference in the amount of silt from each oyster compartment and the amount precipitated in the corresponding shell compartment is the amount of faecal and pseudofaecal matter produced by the oysters in that compartment.

TABLE 15

Amounts of faecal and pseudofaecal matter in cc produced by control (uninfected) and experimental (mostly infected) oysters

Compartment	1	2	3	4	5	6	Total
Control oysters	106	178	198	205	214	107	1008
Experimental oysters	166	130	112	23	154	29	614

The data resulting from this study are presented in Table 15. The control oysters produced 64 percent more faecal and pseudofaecal matter than did the infected group. In all compartments excepting the first the control oysters exceeded the experimental oysters in ciliary activity. Since some of the silt was removed by the oysters in each compartment, the amount decreased successively in each compartment through the trough. However, the efficiency of the oysters in removing the materials apparently increased with the decrease of the amount of suspended silt, for the amount precipitated by ciliary action increased through the first five compartments. Apparently the small amount of silt remaining became a limiting factor when the sixth compartment was reached, for only half as much was precipitated as was the case in compartment 5.

The pattern of precipitation of silt for the experimental oysters was rather irregular. Oysters of compartment number 1 produced the greatest amount, those of compartment number 5 the next largest, and number 4 the least. Thus it is probable that in compartments 1 and 5 (which compare favorably with the controls) uninfected oysters were placed, while in compartments 4 and 6 (in which deposits were negligible) one or more oysters in advanced stages of disease were placed. No experimental compartment attained the average amount produced by the controls.

In the case of the experimental oysters, the erratic variations in amount of faecal and pseudofaecal material produced in the different compartments probably represented variations in disease intensity. According to the sample checked for incidence of the population, one-third were not infected, and another third were lightly infected.

The *second* study testing the effect of *D. marinum* infection on ciliary action in oysters was set up on August 4, 1953, again using the Lund tank, and terminated August 20, 1953. Oysters were raised from spat at the Chene Fleur field station. At the time this study began they were just over two years old. Thirty-eight oysters were selected for approximately uniform size and weighed. They were then matched in pairs of nearly equal weight. One of each pair was used for an "experimental" oyster, and the other was used as a "control". There were 22 pairs thus matched for equal or nearly equal weights. Eighteen pairs were used to initiate the experiment and four pairs were held in reserve as replacements for oysters dying during the study. The number of replace-

ments required was underestimated and replacements could not be made for all oysters that died. Some "control" oysters developed heavy infections and thus became "experimental" oysters.

The oysters in the experimental aquarium were arranged in the order of their weights from the seawater entry end to the exit end of the aquarium. Pseudofaecal and faecal matter was collected and measured for volume. The combined pseudofaecal and faecal matter reflects the ciliary activity of the oysters, and the faecal matter is a measure of the amount of food material actually ingested by the oyster, minus that part digested.

Prior to the starting date of this experiment a sample of 25 oysters from the Chene Fleur group used in this study was analyzed by the thioglycollate method for degree of infection by *Dermocystidium marinum*. The weighted incidence of infection of the sample oysters was 0.48, and 40 percent were found to be infected. There were no heavily infected oysters in the sample.

All experimental oysters were infected with *Dermocystidium* by the "injection" method. Studies described in earlier sections of this report show that this method produces nearly 100 percent infection in one day's time and that multiple infections probably take place, resulting in rapid development of disease to an acute stage. Results of this experiment confirm the efficiency of the infection method used. Since a certain number of experimental oysters were previously infected, the infection by the injection method was superimposed on a previous infection in some of the experimental oysters. It was also added to by natural infection from the aquarium water supply in the course of the experiment.

The "control" oysters in this study were injected with tissue mince of uninfected gapers. These gapers were oysters from Redfish Bay, Louisiana, which died of culling injuries. Three meats of these oysters were broken up in a Waring Blendor, and after dilution with sterile seawater the tissue mince was injected through small holes in the shell into the shell cavity. The control group provided oysters negative for disease, some in light stages of infection.

The disease developed more rapidly in the experimental oysters than was expected and in the 25 days of the experiment, 9 died with heavy infections. Among the controls three died of causes other than *Dermocystidium* disease and three died with heavy infections of *D. marinum*. Some light natural infections must have developed very rapidly in the controls.

When an oyster died in the experimental or control tank it was replaced as long as reserve replacements were available. If the gaper was in the experimental tank, its matched mate in the control was also removed, and vice versa. Only eight oysters remained in each of the control and experimental troughs at the end of the study. All survivors were checked on the closing date for intensity of infection. The oysters were then grouped according to the intensity of infection and the amount of pseudofaecal and faecal matter produced by each oyster was calculated on a per day basis.

A check was made of the water flow through the three troughs. Water level was held constant in an overhead tank by an overflow tube. The glass tubing through which water was fed into the experimental troughs was calibrated for equal diameter in all three. They were equal within very narrow limits and were cleaned daily. The flow was at the rate of one liter per 51 seconds plus or minus one second. Fluctuations were caused by slight fouling of the tubes and varied in the same direction in the three tubes. The flow was timed with a stop watch. The results of this study are summarized in Table 16.

TABLE 16

Amounts of pseudofaecal and faecal matter produced by oysters with different intensities of infection with *D. marinum* (Exp. 1)

Intensity of infection	No. oysters tested	Total no. oyster-days of testing	Faecal matter per oyster per day (cc)	Pseudo-faecal matter per oyster per day (cc)	Total production per oyster per day (cc)
Heavy	8	71	0.48	0.70	1.18
Moderate	6	90	0.62	0.77	1.39
Light	6	72	0.91	1.28	2.19
Very light	4	43	1.01	1.80	2.81
Negative	6	58	1.11	1.70	2.81

The total of pseudofaecal and faecal matter reflected ciliary activity and was indirectly a measure of respiratory activity but probably did not represent, with any great degree of accuracy, the feeding activity of the oysters. Feeding was better represented by the faecal matter production alone. Generally the data in Table 16 plotted as Fig. 17 show infection corresponding inversely with total pseudofaecal and faecal production. The very light and negative classes were combined for this calculation since these two had identical records as shown in Table 16.

The *third* experiment of this series was set up on August 14, 1953, and the study was terminated at the end of a 21 day period. The oysters used in feeding experiment no. 3 were obtained from Milford, Connecticut, through the courtesy of Dr. Victor Loosanoff, Director of the Milford Biological Laboratory of the U. S. Fish and Wildlife Service, and Mr. Joseph Uzmann, guest researcher at that laboratory. A considerable sample of these oysters was checked by the thioglycollate method to determine whether there were any infected ones among them. No infected oysters were found. Experimental oysters were infected with *Dermocystidium* by the injection method, the injections taking place on August 13, 1953. All oysters were weighed and matched in triplets; one of each three was a control and the other two were used in experimental groups A and B. Each of the two experimental groups contained nine oysters and the control group contained nine oysters. The data show that the injection method was successful in producing immediate infection in all experimental oysters. Control oysters picked up a considerable natural infection from the water flow (Table 17); but no control oyster reached a stage of heavy infection in the period of the study, and none died. On the other hand 67.0 percent

FIG. 17. Effect of intensity of disease on ciliary activity as shown by production of faeces and pseudofaeces. For details of experimentation see text.

TABLE 17

Production of pseudofaecal and faecal matter by oysters with different intensities of disease
Period: August 14, 1953 to September 4, 1953 (21 days)

	Weighted incidence of disease	No. oysters used	Oyster days	Pseudofaecal and faecal matter per oysters per day (cc)
Experimental Group A	4.77	9	145	1.95
Experimental Group B	5.00	9	152	1.93
Controls	2.78	9	189	2.68

of the experimental oysters (both groups) died prior to the end of the study (21 days), and on the following day two more died, making a total of 78 percent.

Excepting as noted, the third experiment followed the procedures for experiments 1 and 2 and received the same water flow. Oysters of the third experiment were concentrated in the three compartments of the upper end of the Lund trough. All three divisions were used, the controls in the right side, experiment A in the middle and experiment B on the left.

In assessing the study the following points should be kept in mind. (1) All oysters started out with equal infection. (2) Experimental oysters received a massive initial infection by injection of infective elements into the mantle cavity, and were all infected on the first day. This infection was added to by natural infection from the water stream. (3) The infection intensity increased rapidly in the experimental oysters, ranging probably from light to moderate in the first week, from moderate to moderately heavy or heavy in the second week, and attaining a heavy level in all oysters by the end of the third week. Because of progressive development of intensity from zero to heavy, faecal and pseudofaecal production were predicted to decline from week to week. (4) Control oysters also, with one exception, became infected from the natural water stream. (5) Level of infection in the control group was never as high as in the experimental group, but more than half had attained a level of acute disease (but not heavy) by the end of the third week. (6) With progressive development of disease in the control oysters decreased faecal and pseudofaecal matter were predicted by the end of the third week. (7) The decrease predicted was less than in the experimental oysters, because of the differences in initial levels of infection.

Table 17 contains the data derived from the third experiment on production of faecal and pseudofaecal matter. Experimental groups A and B produced nearly equal quantities of pseudofaecal and faecal matter, much below the level of the controls. Assuming that the controls represent 100 percent, the two experimental groups produced only approximately 71 percent. These data are also presented graphically in Fig. 18.

In computing the values on a weekly basis, all oysters producing for a part or all of each week were included in the calculations. In the case of those producing for only a part of the week, the production figures were increased proportionately to represent a full week. As shown by the graph, ciliary and feeding action were not only considerably less in the experimental oysters, but decreased progressively for the three weeks of the study as infection intensity increased.

The controls showed a different pattern. Ciliary activity and feeding increased in the second week over the first, probably due to acclimatization to the aquarium conditions. In the third week there was a drastic slowing down of ciliary action as the acute disease stage was reached in some control oysters.

Epizootiology[2]

Mackin, Owen, and Collier (1950) first mentioned the relation of temperature and salinity to incidence of *Dermocystidium* in oyster population. Since that time numerous studies have been reported bearing on the development of epizootics caused by *D. marinum*. Mackin and Wray (1952) outlined the basic factors affecting development of epizootics and further discussed the role of physical and chemical factors. Ray (1953a) studied the relation of age of oysters and incidence and intensity of infection. Ray (1954b) and Ray and Chandler (1955) reviewed the data on salinity and temperature as factors in epizootiology and presented experimental data. Owen (1955) conducted considerable experimentation on the effect of low temperatures and low salinity on infection rates, and discussed the relation of field mortality rates and salinity, as well as seasonal variation in incidence. Mackin (1956) presented data on salinity tolerances of oysters and *D. marinum*, Andrews and Hewatt (1957) discussed epizootiology of the parasite and Andrews (1957) studied the effect of density of oyster populations and infection incidence and intensity. Mackin and Sparks (1962) presented data on the development and structure of epizootics on experimental tray populations. All of these authors have presented information in miscellaneous other studies which deal with the problems of epizootiology. These studies will be summarized here.

Composition of Oyster Populations as Related to Disease—definitions.

Recognizable at present, the types of individuals in oyster populations are as follows:

a. *Susceptibles* are those individuals which may contract fungus disease. Degree of susceptibility may vary more or less from individual to individual, and from race to race. It may also vary seasonally in any one individual and possibly may vary with other environmental conditions, or with age of the oyster.

b. *Immunes* are those possessing a capacity to resist infection or development of disease if infected. An immune individual may be a "carrier" if infected but resistant to development of disease. The existence of such individuals, so far as *D. marinum* is concerned, has not been established. Immunity may be acquired or "natural", the latter implying hereditary capacity for coping with disease.

A susceptible (or immune carrier) which has contracted disease is referred to as a "case". There is no way of distinguishing between cases which are infected susceptibles and immune carriers, if the latter exist. The use of the word "case" does not here imply that the oyster is significantly diseased but only that it is infected as shown by the thioglycollate culture diagnosis method.

As considered here, the word "epizootiology" includes consideration of enzootic periods as well as epizootics proper. It considers the effect of disease on populations of oysters but does not exclude examination of data applicable to individuals. What applies to individuals and what applies to populations may be inextricably interdependent.

At present it has not been demonstrated that there are races or strains of *D. marinum* differing in virulence. Although these may some day be proved to exist, it is assumed meanwhile that virulence in *D. marinum* varies with environmental factors but that only one genetic race exists.

Epizootics on Oyster Beds. Studies of mortalities due to *D. marinum* on oyster beds are too incomplete for analysis. It is inferred that because weighted incidences of disease

[2] Epidemiology of an animal population.

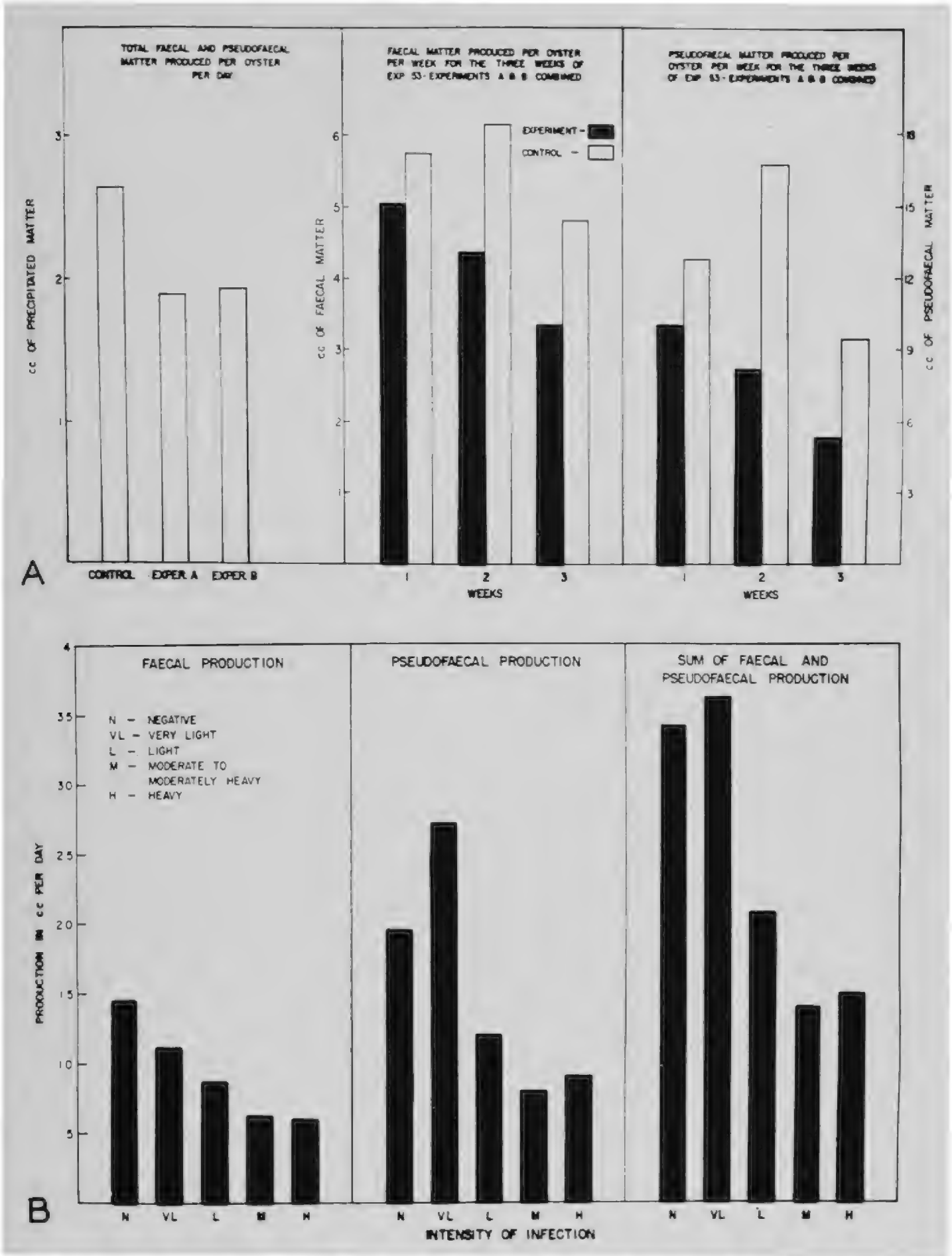

FIG. 18. A. Effect of infection with *D. marinum* on production of faecal and pseudofaecal matter. Experimental oysters were infected by the injection method; controls were uninfected at the beginning of the study, but some became infected from the sea water supply before the study ended. Note the difference in scale of the three graphs. B. Same experiment as shown in 18A, but the oysters were sorted according to intensity of infection as determined at the end of the study.

in oyster populations of planted beds and natural oyster reefs reach figures comparable to those in experimental populations with high mortality rates that similar mortalities occur on the planted beds and reefs. Experimental populations in aquaria with infections equal to those in oyster beds are thought to have generally higher mortalities.

Tray-held populations, on the other hand, are better off than most bottom plantings so far as environmental factors are concerned, and should undergo losses to disease comparable to those in populations of bottom plantings. This belief is strengthened by many observations in the field.

Ordinarily, investigations of reported mortalities are too late for extensive study; the bed is usually found decimated, with large amounts of shell, boxes (hinged shells), and a small number of survivors. The best that can be done under such circumstances is to sample the survivors for intensity of disease. However, there have been a few experimental studies of mortality in planted beds.

St. Amant *et al.* (1958) presented figures on mortalities in experimental bottom plantings. Oysters were planted in November, 1955, when 8 to 12 months old. Approximately 13 months later (Dec. 7, 1956) 82.5 percent were dead. They showed that 50 percent of these died in the summer of 1956 (May to October). They termed these excessive losses "summer mortality" which losses they said "are attributed to the effects of a fungus infection, *Dermocystidium marinum*". Cumulative mortalities as recorded by St. Amant, *et al.*, are presented in Fig. 19. The method of estimation of mortality was not stated, but it is assumed that it was based on use of "boxes". This method is always in error and the actual mortalities are always higher than indicated by the box count method.

Menzel and Hopkins (1952) similarly recorded mortalities in an experimental bottom planting. Their data are as complete as they could be made for bottom plantings. The planting was at Bayou Bas Bleu, where conchs were present only in small numbers. Stone crabs contributed to the mortality along with fungus disease. The authors believed that destruction by stone crabs was higher than could actually be accounted for because of fragmentation of the shells by the crabs and consequent loss of the "evidence". They believed also that *D. marinum* was a major cause of mortality. Mortality in 15 months was calculated to be 75 percent, most of which occurred in summer months with the highest death rates occurring in September and the lowest in January and February. Plantings were also made by Menzel and Hopkins in an area of higher salinity (Bay Ste. Elaine) where conchs played a greater role in destruction of oysters. Fig. 20 shows the form of the mortality curve at this station. There seems to be little difference between the Bayou Bas Bleu and the Bay Ste. Elaine mortality patterns. Both had least mortality in mid-winter and high peaks in the summer after a gradual rise through the spring.

Epizootics in Tray Populations. As indicated above, the bottom planting curves

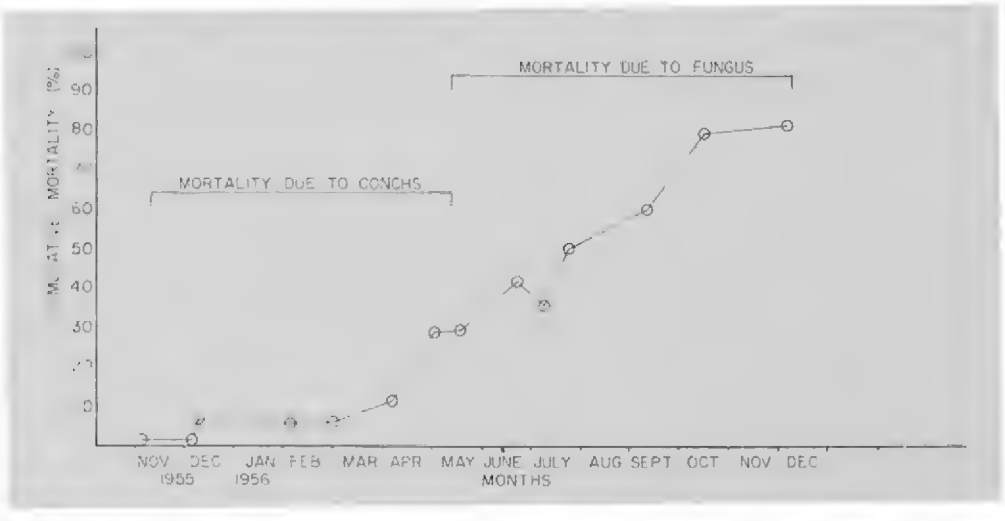

FIG. 19. Mortality of oysters in a bottom planting in Louisiana. Data from St. Amant *et al.* (1958). Mortalities were determined by sampling. Total mortality in a 14 month period was 82.5 percent.

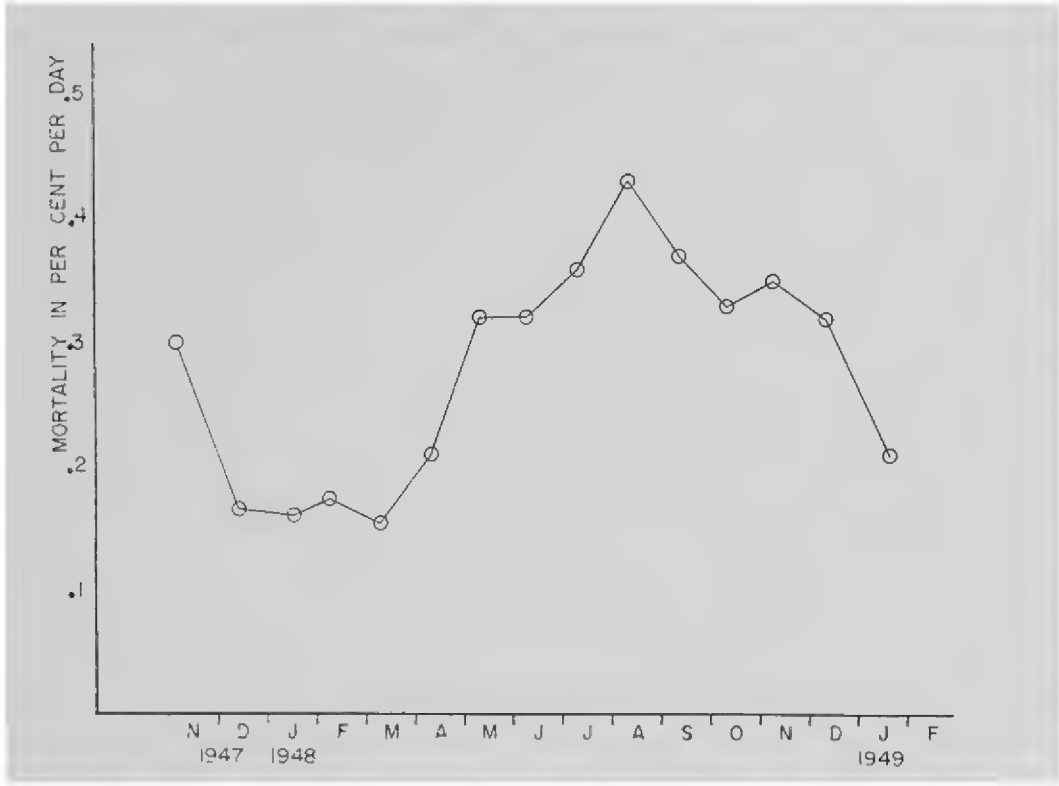

FIG. 20. Mortality of oysters in a bottom planting in Bayou Bas Bleu, Louisiana. Data from Menzel and Hopkins (1952). An undetermined number of the mortalities were due to predators, especially stone crabs (*Menippe mercenaria*). The graph shows a summer peak usually indicative of heavy *D. marinum* mortality.

exhibited herein do not show exact epizootic curves for *Dermocystidium* disease, because predators (and possibly other diseases) were not separable from the fungus disease as a cause of mortality. Experimental populations of oysters held in trays provide much more information because data on incidence and intensity of disease may be obtained along with exact mortality rates. Predators can be largely eliminated as mortality producers by caging the oysters, and morbidity data as well as mortality data from fungus disease are available from periodic samplings. In those areas easily accessible, so that examinations can be made daily or weekly, dying oysters may be recovered in sufficient percentage to make relatively certain what part of the total deaths are due to disease and what part to other causes. Lastly, experimental populations held in trays may be selected for intensity of disease when experimentation begins, and data on rate of infection of uninfected populations may then be obtained.

Andrews and Hewatt (1957) measured the impact of fungus disease on oysters held in trays in the York River estuary in Virginia. Oysters from areas free of the disease (James River seed grounds) were moved to an area where the disease is enzootic. Epizootics resulting were measured in the summer of 1953 and the same population was observed through the winter and to the end of 1954. These 800 oysters were "planted" in April of 1953 but remained free of disease until August when mortality from fungus disease began. A small epizootic resulted and reached a peak in death rates in September. About 9 percent of the population died during August, September, October, and November. The records show that more than half the population became infected, but the infection peak was reached in late October or early November. By the following March, 1954, the disease had disappeared from the population (had dropped to such a low ebb that the standard culture method of diagnosis failed to reveal its presence). Infections in the second year appeared in June, two months earlier than in the previous year. A massive epizootic developed during the summer, resulting in loss of about 64 percent of the remaining population. The peak of this epizootic was August and September, and deaths had ceased to occur by December. Fig. 21 shows the trend of epizootic develop-

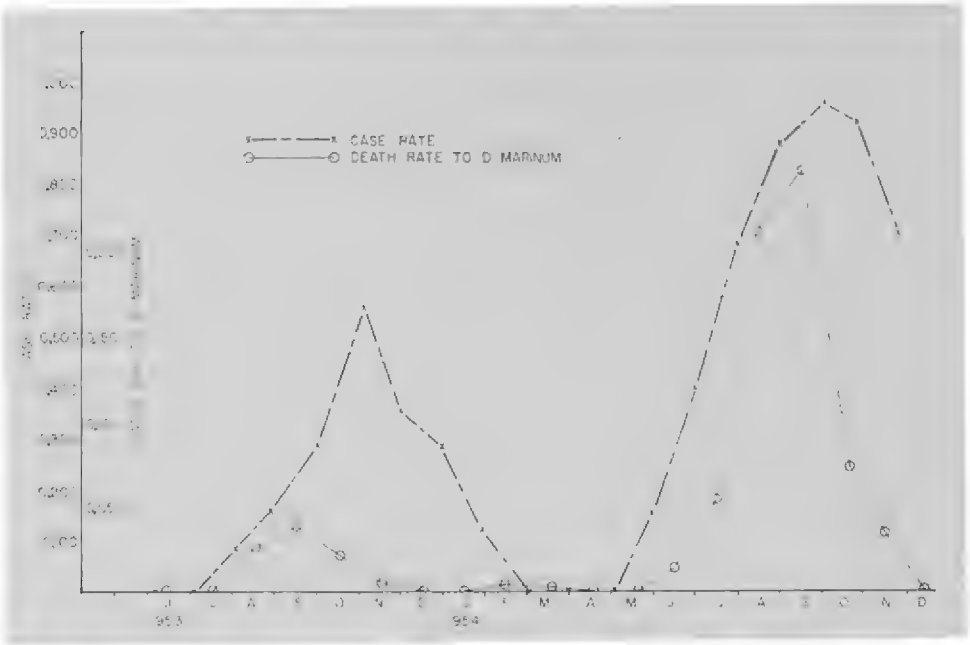

FIG. 21. Case rates and death rates over a two-year period in oysters held in trays in the York River. Data are from Andrews and Hewatt (1957). Oysters were free of disease at the beginning of the study. Although more than one-half of the oysters became infected in the first summer, heavy mortality from disease did not occur until the second summer.

ment in this tray population. Case rates, total death rates, and rates of death due to *D. marinum* are shown. The lag in elimination of infections after all deaths had ceased is particularly well illustrated in these graphs. Also, in the epizootic of 1953, fungus disease accounted for only about 60 percent of all deaths, while in 1954 fungus disease accounted for 95 percent of all deaths. During the peak of the 1954 epizootic, there were hardly any deaths due to other causes, and by October 96 percent of the population was infected. It is interesting that small mortalities developed in February and May of 1954 which probably indicated the presence of a minor disease other than the dominant fungus disease.

Another tray population studied by Andrews and Hewatt showed a similar epizootic development (Fig. 22). Earliest infections occurred in April (not shown in the graph). In May a scattering of deaths occurred and the epizootic reached its peak in September. Fifty-two percent of the population died of fungus disease in the period from May to

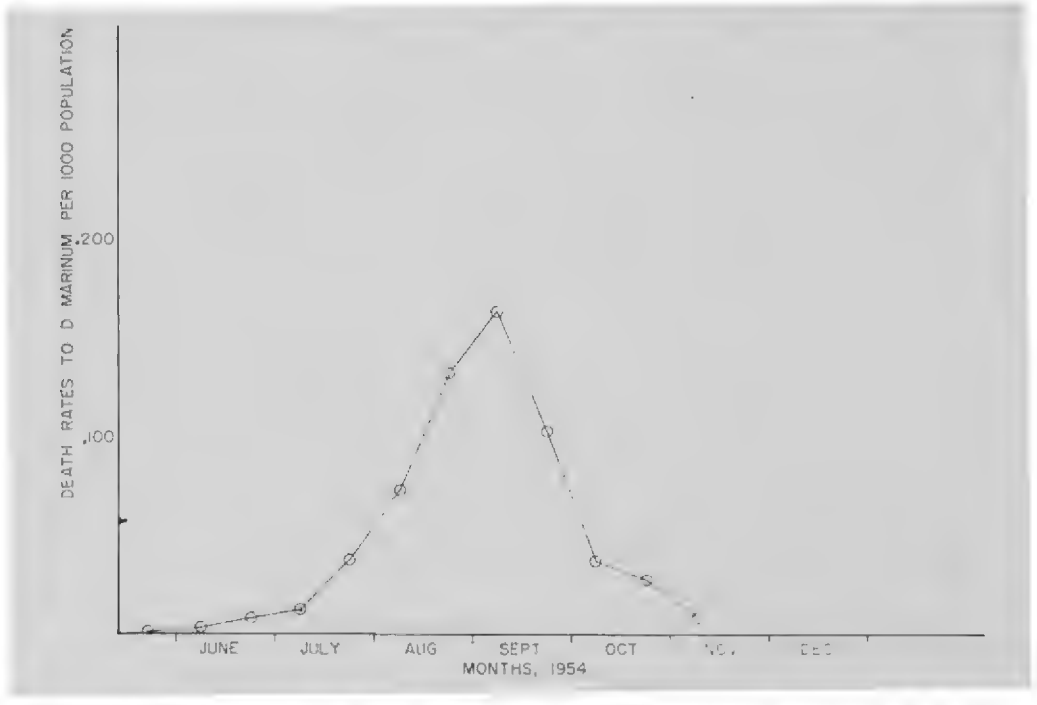

FIG. 22. Death rates of oysters due to *D. marinum* in the summer and early fall in Virginia. Data from Andrews and Hewatt (1957). This population was taken from endemic waters and held in trays in the York River. Fifty-two percent of the oysters died of *D. marinum* infection in this epizootic.

November, which was 87 percent of all deaths. These oysters differed from those of the population shown in Fig. 21 in that they were natives which had always been in enzootic areas.

At the northern limit of the range of *D. marinum* in Delaware Bay, minor epizootics are initiated each year in the period between May and August. The peak of incidence is in September or October (Anonymous, 1958). It is believed by New Jersey investigators that annual mortality rates due to the fungus average only about 5 percent, with a possible maximum of less than 20 percent in the heavily infected areas of "The Cove". The rise of incidence is later in the summer than is the case in the lower Chesapeake and reaches its limited peak just prior to onset of cold weather. The period of high temperature is too short for full development of epizootics of the type found so destructive in Louisiana. Complete disappearance of the disease takes place in winter or early spring (based on culture tests).

In Louisiana the epizootics usually have their inception in April or May. The rapidity of development varies with environmental conditions, and with number of susceptibles per unit area. The disease in heavily enzootic areas never disappears in winter and limited epizootics may develop in unseasonably warm periods in winter. The form of epizootics is often more variable than in the Chesapeake or in Delaware Bay.

Mackin and Sparks (see last paper, this volume, 1962) studied the development of epizootics at seven stations in Barataria Bay, Lake Grande Ecaille, and Billet Bay. Tray populations of 1000 oysters were placed at each station. Monthly death rates and death rates due to *D. marinum*, averaged from all seven stations, are shown in Fig. 23. The hump occurring in May on the curve of total deaths was associated with "mycelial disease" (see below). In high salinity waters of Louisiana, spring and early summer epizootic peaks may occur which coincide with the spring tide peak; this peak usually does not occur in low salinity areas. Mean cumulative mortality in the seventh-month period shown in Fig. 23 was about 60 percent for the seven stations, and the range was from about 30 percent in the station with least mortality to 80 percent in the station developing the most severe epizootic.

The percent of the total deaths attributed to *D. marinum* progressively increased from

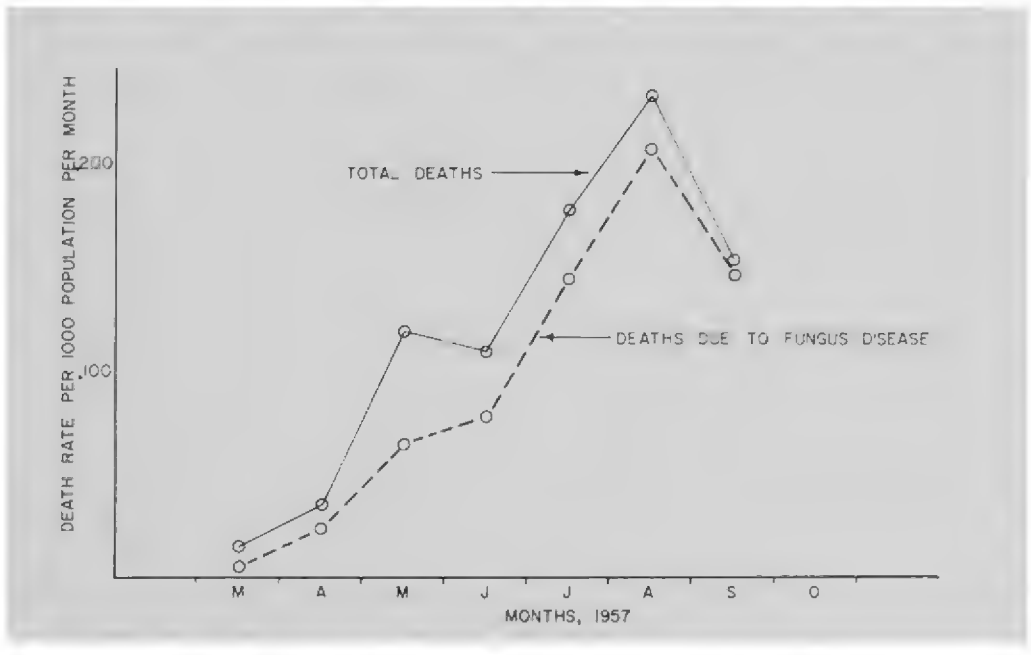

FIG. 23. Death rates, and death rates of oysters due to *D. marinum*. Oysters were held in trays at seven stations in Barataria Bay, Billet Bay, and Lake Grande Ecaille, Louisiana, 1000 oysters at each station. Data are averages for all stations. Data from Mackin and Sparks (this volume, 1962). Cumulative mortality was about 60 percent.

early spring to September. At the same seven stations, the increase was from 26 percent in March to 100 percent in September (Fig. 24).

Experimental Epizootics in Aquaria. Epizootics initiated in aquaria by artificial infection methods culminate rapidly, and run their courses in much less time than the epizootics observed in field trays and bottom plantings. Infection by the "injection" method (see section on pathogenicity) produces especially sharp epizootic peaks. The progress is limited only by the number of available susceptibles and comes to an end when the population is destroyed. The developmental sequence of the epizootic is distorted by the high concentration of infective cells, and by the simultaneous infection of all individuals in the experimental populations. In Fig. 25 is shown the form of such an epizootic. The scatter of points at the end of the study indicates that the population was so decimated that the data came to be based on statistically unreliable numbers of survivors. The first oyster died on the tenth day and had developed an advanced case of disease, but not the heaviest type. On the fifteenth day two oysters died with heavy disease. Only four oysters survived beyond the fortieth day and all of these were in advanced stages of disease.

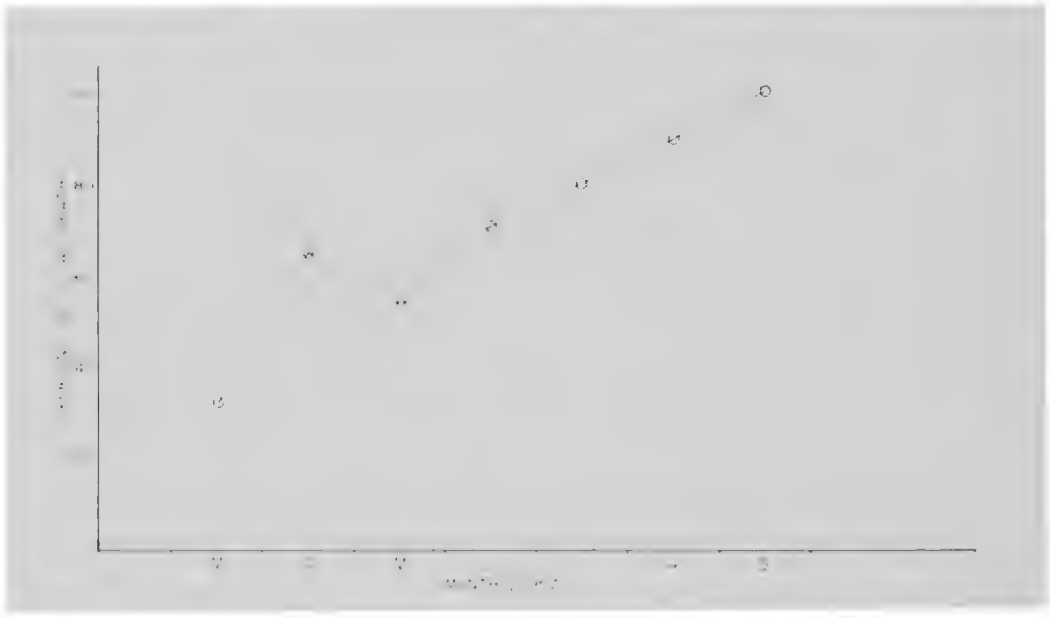

FIG. 24. Percentage of total deaths of oysters due to *D. marinum* in different months; 7,000 oysters were tested at seven stations in Louisiana. Data from Mackin and Sparks (this volume, 1962). The increasing deaths are correlated with temperature.

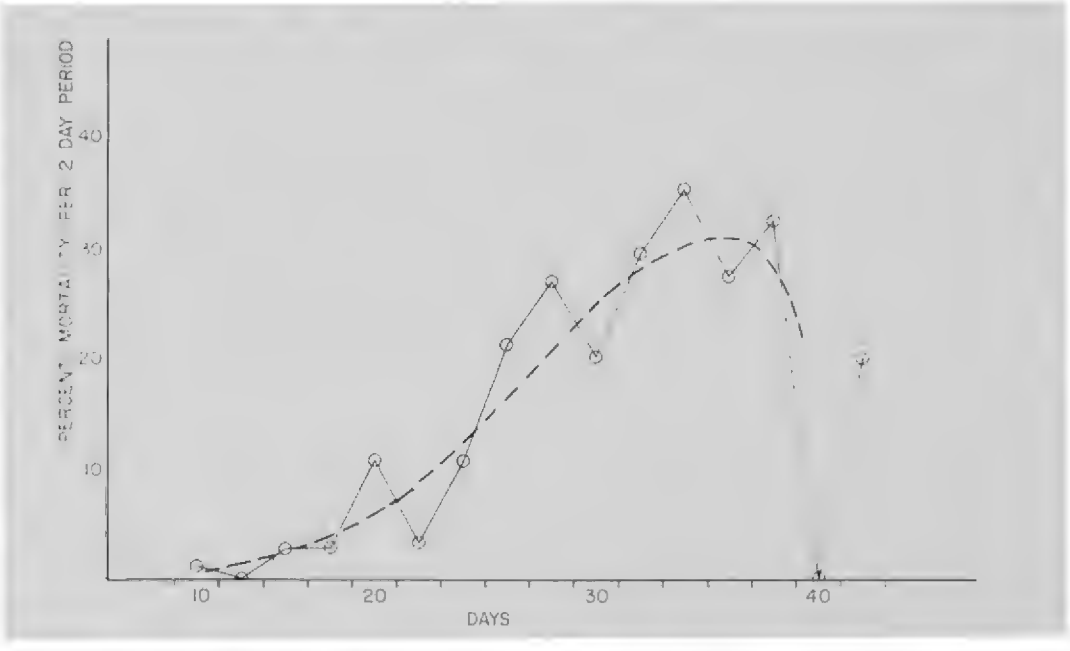

FIG. 25. Experimental epizootic in aquaria. Oysters were "injected" with infective cells and the first oyster died with advanced disease on the tenth day. The population was virtually wiped out by the forty-second day.

In another set of experiments in aquaria (Mackin, Ray and Boswell, 1953), oysters were "fed" the infective cells. By this method, infections are not simultaneous but take place over a period of several days, and initial infection dosage is small. The first oyster died with heavy disease on the sixteenth day and the epizootic developed in an accelerating manner as more oysters died and liberated infective cells. In this type of aquarium epizootic 63 days were required to produce a 93 percent mortality, about twice as long a period as in the cases where the infection method produced disease in all individuals of the population simultaneously. The form of this epizootic is shown in Fig. 26.

Biological Factors in Epizootiology. Several biological factors affect death rates from fungus disease and vary the development of epizootics. These will be discussed in the next sections.

Infection and Invasion of the Oyster Host. Portal of entry of a parasite may be an important factor in epizootiology. The author (1951a) believed that infective cells were ingested with food, phagocytized in the stomach and carried into the epithelium of the digestive tract of the host by the phagocytes. This still seems to be the best hypothesis, but its acceptance does not rule out the possibility that there may be other portals of entry. Ray (1954b), and Ray and Chandler (1955) excised small pieces of gill and mantle tissue and were able to induce infection in these tissues by placing them in vials containing large numbers of *D. marinum* cells. The author repeated these studies and confirmed the entry of infective cells into the isolated tissues. It is believed that this is not a usual portal under natural conditions due to the tendency of phagocytes to remove foreign cells from external epithelial surfaces.

The tendency for phagocytic cells of oysters to engulf foreign cells and substances (Stauber, 1950) and to migrate through tissues makes it possible that any stage of *D. marinum* may become an infective cell. All data so far seem to support this hypothesis. However, since it has not been possible to separate the different cell types in infection experiments, the question has not been settled. Culture studies (see Section II) indicate that cells of *D. marinum* freed from the host may undergo continued reproduction and development in seawater. Cultures all tend to end with production of a type of non-motile spore, and it may be that this spore is the cell initiating epizootics in spring and summer after the apparent disappearance of infections in winter (in the

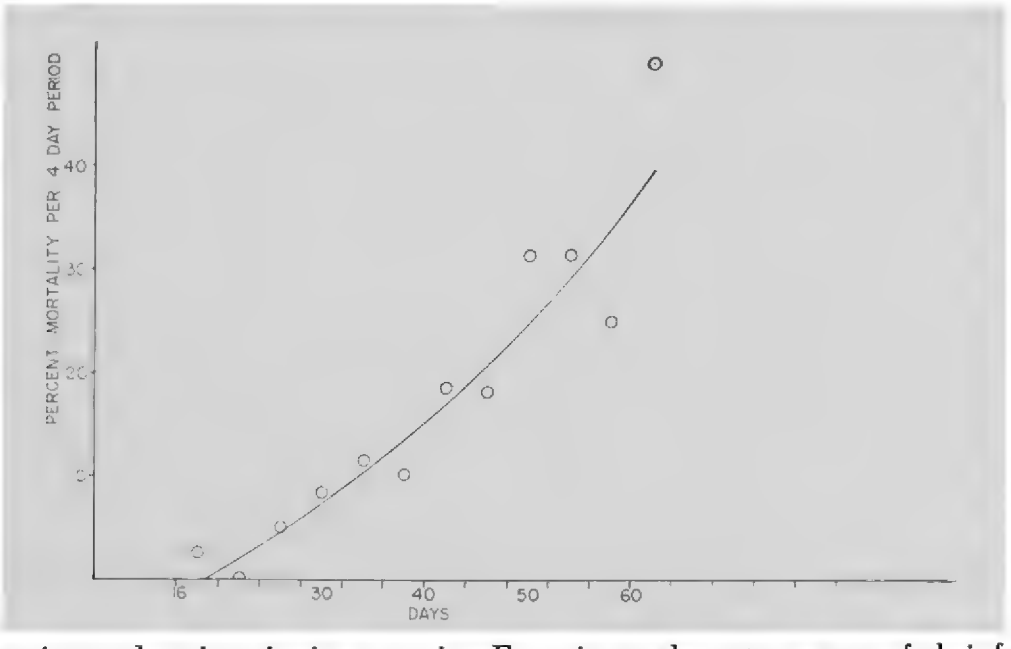

FIG. 26. Experimental epizootic in aquaria. Experimental oysters were fed infective cells of *D. marinum*. Sixty-three days were required to produce a 93 percent mortality.

northerly part of the range of distribution). It is highly unlikely that cells of *D. marinum* in sea water can survive cold in all stages. The hypnospore is probably the usual over-wintering stage; it is likely that with the beginning of spring, this cell produces sporangia from which come the infective cells.

Andrews and Hewatt (1957) conducted studies to determine whether or not oysters may carry over-wintering stages, which produce the initial "infection" internally in the following spring. They used oysters from enzootic areas, but from which the infections had disappeared during the winter. These were placed in closed aquaria at 23° to 28°C. Although the thioglycollate tests failed to reveal any infections in these oyster populations at the start of the study, some oysters developed typical cases in the warm water and infected other oysters. It is not certain that this shows a special infective stage unresponsive to the thioglycollate culture diagnosis test. It may mean that when infections are few, and are extremely light when they do occur, the thioglycollate test will detect the presence of the parasite only if much larger numbers of oysters are used than is usual.

Andrews (1956a) infected oysters by keeping them in an endemic area. These oysters apparently lost their infections in the winter. In the spring they were transferred to low salinity areas where *D. marinum* does not occur. Although thioglycollate tests showed no infections when the oysters were moved, a few nevertheless developed infections in the following summer. This is taken to indicate that some infections do overwinter in Virginia and contribute to or originate epidemics in the summer-fall period.

Whether or not oysters may harbor latent infective stages, it seems certain that populations may be infected from external sources, after periods of complete freedom from infection. Oysters from various stations in Redfish Bay, Louisiana, in 1953 were completely free from infection, as shown by repeated thioglycollate culture tests in warm weather (Mackin, 1954a). Oysters from the same bay were found to have acquired a low level infection one year later. It is believed that these infections came from a distant source, since oysters from all known reefs in the Bay and "outside" in the Gulf were tested in 1953 with negative results.

Invasion and distribution of infective cells within the body of the host appear to present no problems. Phagocytic cells move freely through epithelia and connective tissue, and blood sinuses and vessels may introduce parasites to all tissues. No tissue has been found to be resistant to attack, except possibly the renal epithelium.

In a sense, there are two types of epizootics caused by *D. marinum*. One involves populations of oysters, the other populations of host cells after invasion of a host oyster is accomplished. Since *D. marinum* is essentially an intracellular parasite, an individual host cell is also a host in itself. Movement from host cell to host cell is accomplished only when a host cell dies and distintegrates, liberating the contained infective cells into the blood sinuses. Some cells of oysters are phagocytic even when they are components of tissues other than the free blood cells proper. Infections of these cells take place readily. Other cells seem to be actively penetrated by the parasites, by means of a lytic process, and cells of *D. marinum* may develop in intercellular spaces, such as the gonadal cavity, when in close contact with host cells.

Defense Mechanisms. So far as is known, a combination of phagocytosis and diapedesis is the only effective defense of the oyster against infection. By this process, parasitic cells are picked up in the tissues, and carried across epithelia into the gut lumen or to

the exterior. Phagocytic cells are shed in large numbers by oysters in acute stages of disease until breakdown of phagocyte production occurs. The method of demonstration of diapedesis is to place starved oysters on a black background. Some hillocks of phagocytes on the black substrate are clearly shown by this technique and may be shown to contain parasites by smear or thioglycollation.

Diapedesis is not usually effective in ridding oysters of *Dermocystidium* in warm months, when the parasites are multiplying rapidly. It is believed that oysters are able to free themselves of these parasites in winter, when reproduction of the parasite ceases, if intensity of infection is not too great. In those estuarine areas where the number of infective cells is low, the phagocytes may be able to rid an oyster of isolated infection even in warm months. It has been observed that oysters infected in early spring may rid themselves of parasites in the summer following if extensive flushing of the estuary occurs. Ray (1959) found that in Bayou Dau Haute (Louisiana) 12 samples of oysters taken April 23, 1959, all showed infections. The weighted incidence of the 12 averaged 0.36 and the incidence ranged from 20 to 60 percent. In the same bayou on July 15 of the same year, 10 samples out of 11 taken were free of the parasite and in the 11th sample only one very lightly infected oyster was found. The weighted incidence of all oysters in all samples on July 15 was 0.009. This decrease followed heavy local rains, and flushing freshets in the bayous.

Age of the Host and Infection Rates. Mackin (1951b) reported that young oysters, less than one year old, appeared to possess an immunity of some kind. For many years it has been known that the excessively high mortalities occurring in Louisiana oysters were primarily among the "counter" oysters. These oysters must remain on beds for a longer period than do oysters raised for canning, and hence are older when harvested. Ray (1953) made a study of infection incidence and intensity in oysters less than one year old, compared with those older than one year, in Louisiana. He caught spat in the middle of June, 1952, in shell bags. These spat were marked and hung in wire bags from stations in Barataria Bay. Other spat of the same age were taken on shells at the laboratory in Bayou Rigaud. Oysters known to be 11 to 12 months old from Bay Chene Fleur were compared with the spat. These year-old oysters at the start of the study had only a very low incidence of *D. marinum*. A sample checked by the thioglycollate method at the start of the study showed all negative, but it was known that a very low residual infection was present at Chene Fleur. Another group of oysters about four years old from Gardiner's Bay, Long Island, New York, was also used in the study. This group of oysters was negative for disease at the start. These groups of oysters were observed for intensity of infection for about a year. The thioglycollate method of diagnosis was used. Samples were taken at irregular intervals. The results of Ray's studies are summarized in Table 18.

A study of the table shows that a very light infection appeared in one spat group about five weeks after the study started, and in the other about eight weeks after the start. These spat were six to eight weeks and nine to ten weeks old, respectively, when the infections appeared. However, no infections heavy enough to be considered "disease" appeared in either group of spat until they were approximately a year old. Even then the intensities and incidence of infection were very low compared to those of older oysters.

The year-old Louisiana native oysters became infected in four weeks and in five weeks

TABLE 18

Speed and intensity of infection of young oysters compared with older oysters. Data from Ray, 1953

Weeks from start	Spat group A		Spat group B		1-year-olds from Chene Fleur		4-year-olds from Gardiner's Bay	
	W.I.*	I.†	W.I.	I.	W.I.	I.	W.I.	I.
0	0.00	0.0	0.00	0.0	0.00	0.0	0.00	0.0
1	0.00	0.0			0.00	0.0	0.00	0.0
2			0.00	0.0	0.00	0.0		
3	0.00	0.0			0.00	0.0	0.15	20.0
4					0.15	30.0	0.25	30.0
5	0.00	0.0	0.08	3.8	0.35	50.0	0.10	20.0
6							1.55	70.0
7							1.35	60.0
8	0.04	3.7			0.65	40.0	1.20	60.0
9							2.00	60.0
11	0.00	0.0						
12					1.00	60.0		
14							0.35	50.0
15					0.53	60.0		
16							0.50	50.0
22	0.07	6.6						
27.5							2.55	80.0
31	0.13	6.6						
36.5							2.30	90.0
40	0.27	13.3						
50	0.66	37.0						
51			0.50	26.3				

* W.I. = Weighted incidence.
† I. = Incidence, percent.

had attained a level of infection comparable to that of the two spat groups a year later. At twelve weeks the year-old oysters had reached 60 percent infection and weighted incidence of 1.00.

The four-year-old oysters became infected at three weeks and reached a high level of infection of 70 percent and weighted incidence of 1.55 in six weeks. A very rapid acceleration of incidence rates and intensity of disease occurred between the fourth and sixth weeks, and heavy mortality occurred. Another group of Long Island Sound oysters showed similar excessive acceleration of infection in the early weeks of sojourn in enzootic waters. It is assumed that these oyster populations were made up largely of highly susceptible individuals, and probably were not properly comparable to the Louisiana oysters.

Andrews and Hewatt (1957) showed that young oysters, when fed tissue mince of heavily infected gapers, were as easily infected as were older control oysters and died as rapidly. These authors believed that young oysters fail, under natural conditions, to ingest enough infective cells to produce disease and that the "immunity" of immature oysters is due primarily to failure to ingest the minimum infective number of spores. Their hypothesis is supported by the author's studies of the effect of varying dosage of infective cells, reported in the section dealing with pathogenicity. There is some evidence that infection rates increase up to about four years.

Density of Oyster Populations and Infection Rates. Increasing the number of oysters per unit area, while the percentage of susceptibles in the population remains constant, has the same effect in increasing infection rates as increasing the percentage of susceptibles in the population when the area and the population remain constant. Density

of oyster plantings, therefore, must necessarily affect infection rates, and as a corollary, degree of isolation of plantings is also important.

Andrews (1957) mixed infected oysters with uninfected oysters in a tray. The uninfected oysters in this tray developed significantly higher incidence of disease than did a control tray 40 feet away which contained only uninfected oysters to begin with. Andrews concluded from this that isolation of plantings, thorough cleaning of beds, and fallowing would materially delay development of disease in oyster bedding grounds.

The author found that a tray of oysters with more than fifty percent infected developed a summer epizootic of less intensity than oysters in a tray 20 feet distant which started out with only uninfected oysters (Fig. 27). The uninfected tray contained highly susceptible oysters. Andrews' conclusions appear to be correct only when susceptibility is constant. On the basis of other observations, Andrews believed that reduction of number of infective cells with distance was so great that rapid infection from outside sources could be considered negligible. The author does not believe that this is always true. There are many cases of isolated populations of Louisiana oysters where infection took place over what appeared to be prohibitive distances. The case of oysters in Redfish Bay is illustrative. In 1953 a thorough check of several stations showed no infections in the entire bay. Oysters were tested in the laboratory under summer temperature conditions and failed to reveal latent infections. Yet in April of the following year significant infections of oyster populations had appeared in Redfish Bay. There was no apparent local source of infective elements in the Bay or in the Gulf. Redfish Bay is at the extreme tip of the active Delta of the Mississippi, and a minimum of 15 miles of fresh water separated the bay from any known source of infection, unless there is a source in the open Gulf. But oysters 1000 feet in the Gulf off Redfish Bay showed no infections in 1953. It must be concluded that transport of infective cells may take place over long distances of open water barren of oysters.

However, given a nucleus of infected oysters in a population, density of an oyster population controls to a great extent the progress of epizootics. In Louisiana, seed oysters nearly always include some oysters two to three years old. A normal planting is 400 to 600 sacks per acre. Such plantings contain about 8 to 12 oysters per square foot. Such

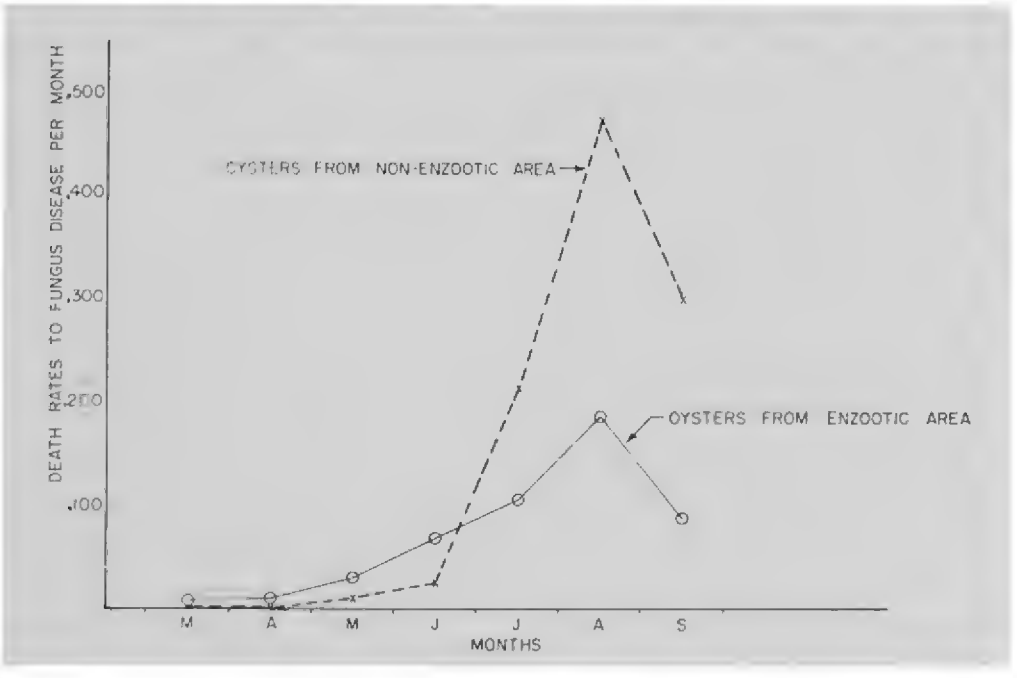

FIG. 27. Effect of introduction of susceptible oysters into an endemic area. Data from Mackin and Sparks (this volume, 1962). One thousand disease-free oysters from a non-endemic area were compared with a similar number from an endemic area. Infection rate was above 50 percent in the latter group at the start of the study, in Barataria Bay, Louisiana.

densities must contribute greatly to the initiation of summer epizootics. Producing counter oysters in Louisiana may depend on natural reduction of plantings through disease or other factors to the point where transmission is slowed appreciably. Much study remains to be done in this field.

Dead and Live Oysters as Sources of Infection. Although infected live oysters undoubtedly contribute to infection of other oysters, oysters dead of *D. marinum* infection contribute materially more. Dead oysters are shredded by crabs and small fishes, liberating uncounted infective cells into the water. Here the classic concept, that a successful parasite is one so adjusted that infection does not result in death of the host, does not apply. *D. marinum* is a highly successful parasite partly because it often destroys its host. This is true of many fungus diseases. In the case of the oyster-*Dermocystidium* relationship, the oyster host is successful because of the "immunity" of young oysters, which in Louisiana reproduce within one to several months after setting (Menzel, 1951a). These young oysters produce extensive natural populations which flourish until their second summer. *D. marinum,* because of its selectivity for older oysters, merely limits longevity of the host instead of destroying the entire population. The oyster population is in no danger of extinction, except locally where certain predators (*Thais* and *Stylochus*) attack the young population while fungus disease destroys those older oysters that escape the predators. The old eroded dead oyster reefs of the Gulf bays all lie in areas where both predators and *Dermocystidium* kill oysters. Extensive seed beds occur where *D. marinum* alone causes most mortality. Most seed oysters are (in Louisiana) about one year old, not yet old enough to have much mortality from *Dermocystidium* disease.

Spawning and Disease. It was observed early in the investigation that heavy mortality of oysters often follows spawning. It was assumed that in some manner spawning weakened the host oysters so much that they were more easily infected and had less resistance to development of severe disease. This matter was studied (Mackin, 1953) in 1950 and 1951. Large numbers of oysters were sectioned and stained before, during, and after the spawning period in both years. It was assumed that if the increase in mortality following spawning was due to *D. marinum* infection, significantly more of those oysters, shown by the sections to be spawned out would be in advanced stages of disease than would others. No such relation was found. Neither was it found that advanced sexual development was related to incidence of disease. It was determined that acute disease during the period when gonadal development should take place (March and April) effected castration of the host. As high as 15 percent of the live oysters studied showed the castration effect and in June, July, and August, 9 to 34 percent of the gapers dying of fungus disease showed degenerating gonads (in Barataria Bay). The sections showed that degeneration of the gonad occurs only when disease strikes in early stages of gonadal development. When infection occurred after gonadal development was materially advanced, no effect on the gonad was observed.

Racial Immunity and Racial Susceptibility. Andrews and Hewatt(1957) compared oysters from different regions and found that those from South Carolina had significantly lower infection and death rates than did other oysters. These oysters were placed in trays in the York River when they were spat size, *i.e.,* only a few weeks old. When compared with Virginia natives they showed consistently lower death rates and infection rates through three years. Part of their data are reproduced in Table 19.

The data indicate that South Carolina oysters showed more resistance to infection than

TABLE 19

Comparison of infection rates and weighted incidence of fungus disease in South Carolina and York River oysters. Data from Andrews and Hewatt, 1957

Origin of oysters		Age, years		
		1	2	3
South Carolina	Weighted incidence	0.10	0.20	0.42
	Incidence, percent	10.0	20.0	33.0
York River	Weighted incidence	0.0	1.56	1.20
	Incidence, percent	0.0	76.0	80.0

did the natives. The data in Table 19 include only those comparing the two populations in the same year. Andrews and Hewatt included other data which compared the same ages in different years. When all the data are carefully studied, it appears that the South Carolina oysters consistently had lower incidences and lower weighted incidences, in their second and third years. It is highly desirable to find resistant races of oysters; the continued existence of the oyster industry may depend on this.

Oysters from the Virginia seaside on the other hand appeared to be more readily infected, and developed higher weighted incidences and death rates, than did York River natives. It is not certain, however, that this indicates a lesser resistance. *D. marinum* is not present on the Virginia seaside, and it appears that the greater susceptibility found by Andrews and Hewatt merely reflected the inclusion of greater numbers of susceptibles in the population. After these were weeded out in the first year in enzootic waters, their susceptibility paralleled that of James River natives. Both of these populations appeared to be highly susceptible, and it is believed that the absence of the disease in the area from whence the oysters were taken is the controlling factor. Such populations are composed almost entirely of susceptibles. Andrews and Hewatt's data are difficult to interpret because the year 1954 was one of exceptional environmental suitability for the fungus and most populations showed significant increases in infection rates as compared with 1953. For example, James River two-year-olds in 1953 had a weighted incidence of 0.78, but the same age oysters in 1954 had a weighted incidence of 1.40.

The effect of large numbers of susceptibles in oyster populations, when they are introduced into enzootic waters, is illustrated by the study by Mackin and Sparks (last paper, this volume, 1962). They introduced oysters from a non-enzootic area to lower Barataria Bay and compared development of the epizootic produced with that of oysters from an enzootic area that already had a high incidence of disease. Oysters of both groups were introduced into the southern Barataria area in early March and kept in trays. After an initial slow start, the oysters from the non-enzootic area developed a severe epizootic which nearly destroyed the population in one summer. The graph, Fig. 27, compares the death rates from fungus disease in the two populations. Trays holding the two populations were only 20 feet apart, but tidal currents did not run from one to the other, although wind-driven currents may have done so at times. The close proximity of the stations was chosen to produce, as exactly as possible, the same environmental conditions in the two populations.

This is a case in which a population with a long head start in infection, nevertheless failed to develop as much of an epizootic as oysters that started from zero level. It

indicates the danger of introducing susceptible populations from non-enzootic areas into areas with high levels of disease.

Host Specificity. Ray (1954b) attempted to transmit *D. marinum* to *Venus mercenaria* and *Mya arenaria.* He also attempted to transmit *Dermocystidium sp.* from *Macoma balthica* to these hosts and to oysters (*Crassostrea virginica*). Although he succeeded in establishing some parasites in molluscs other than their natural hosts for short periods, he believed that these were not suitable as hosts. He considered that all of his attempts to cross-infect were failures, and concluded that species of *Dermocystidium* possess a fairly rigid host specificity.

Ostrea equestris apparently may become infected with *D. marinum* as may *O. frons* and *O. lurida* (Ray, 1954b). Although it was assumed by Ray that the species of *Dermocystidium* found infecting *O. equestris* at Grand Isle was *D. marinum,* it is not certain that this is true. However, the author has studied the *Dermocystidium* in *O. equestris,* and its morphology appears to be identical with that of *D. marinum.* Since Ray (1954b) reported that Uzman infected *O. lurida* with *D. marinum* from *C. virginica,* it appears most likely that species of both genera of oysters may serve as hosts to *D. marinum.*

Temperature and Seasonal Aspects of Epizootics. It has been shown that epizootics produced by *D. marinum* appear mostly in summer. St. Amant, *et al.* (1958) refer to these epizootic waves as "summer mortalities." All data to date show that temperature is the most important of all factors in stimulating epizootic waves and in controlling these waves. The range limit of *D. marinum,* at least at the northern end, appears to be controlled by temperature. The southern limit also may be controlled by temperature, but this is not known to be true.

Mackin, Owen and Collier (1950) first mentioned that low temperature controlled development of *D. marinum* infections, but did not present specific data. Mackin (1948) studied the effect of cooled water in controlling death rate in oysters at Grand Isle. Experimental oysters were placed in a large wooden aquarium, and by means of refrigeration equipment the water flow was kept at a temperature of 17.5 to 18.3°C. Control oysters were kept in a similar aquarium and the temperature allowed to fluctuate with that of the natural water flow, which ranged from 18 to 30°C. The low temperature of 18°C occurred on only one day (during a hurricane). Otherwise the control tank temperature rarely fell below 25°C. Twenty-five oysters were used in each tank and the study was terminated at the end of seven weeks when the control tank reached a mortality rate of 60 percent. Mortality data from this study are presented in Table 20.

Survivors were checked for glycogen content and the results compared with pre-experiment measurement of a sample from the same population. The oysters in warm water lost 72 percent of their glycogen during the experiment while the oysters in cooled water gained 11 percent and in addition gained in total wet weight.

TABLE 20

Mortalities in cooled water compared with death rates in a warm water aquarium
Aug. 10, 1948 to Oct. 2, 1948

Tank	No. oysters	No. dead	Percent dead
Cool water	25	1	4
Warm water	25	15	60

Mackin (1951c, 1953a) repeated this study with refinements. Measurements were made of the incidence and weighted incidence of *D. marinum* in all gapers and survivors in the experimental tank (cooled) and the control tank (warm water). Data from this study are presented in Table 21.

Although weighted incidence of oysters in the warm water nearly doubled that of those in the cooled aquaria the incidence of infection in the two aquaria was equal (60 percent). It appears that the temperature of the water in the cooled aquarium was not sufficient to reduce infection rates in the period of the study but arrested development of the fungus.

Owen (1955) conducted a series of studies of the effect of temperature on death of oysters infected with *D. marinum*. The studies were carried out in winter and compared winter mortalities in aquaria with artificially-produced summer temperatures, and contrasted effects of temperature on oysters with high incidence and low incidence of fungus disease (Table 22). Owen called his low incidence oysters "uninfected." However, he used the sectioning method of diagnosis which is not efficient in detecting low intensities of infection. Owen got his "uninfected" oysters from Chene Fleur Bay, Louisiana, and Cedar Point Reef in Mobile Bay, Alabama. Thioglycollate checks have shown that oysters in Chene Fleur Bay usually have a low level of infection, and the Cedar Point populations are likewise infected, sometimes heavily infected. Owen's "uninfected" oysters are here designated as "low-level-infection" oysters.

Owen's results showed again that oysters held in water of high temperature had high death rates and that the oysters dying were all infected. Owen did not measure the intensity of infection in some of his experimental oysters, but his data showed that most of those dying were heavily infected. His data also showed that oysters which had relatively low incidence of infection or in which there was no infection at all were not ad-

TABLE 21

Study of the effect of cooled water in controlling death rate to *D. marinum* in Louisiana oysters May 18, 1950 to July 19, 1950

Tank	Temperature	No. oysters used	No. dead	Percent dead	Weighted incidence, gapers and survivors
Cooled	18° ± 1°	25	2	8	1.2
Warm	25° to 30°	25	12	48	2.2

TABLE 22

Summary of results of studies by Owen (1955) on effects of low and high temperature on mortality from fungus infection

Study no.	Dates	Tank	Temperature regime	Incidence of *D. marinum*	No. of oysters	Average temp. °C	No. dead	Percent dead	Percent infected by *D. marinum*
1	Dec. 12, 1949	A	Warm	High	20	27.1	10	50	100
	to Jan. 21, 1950	B	Warm	High	10	27.1	7	70	100
		C	Warm	Low	10	27.1	2	20	100
		D	Cool	High	20	15.0	1	5	... ..
		E	Cool	High	10	15.0	1	10	100
		F	Cool	Low	10	15.0	0	0	
2	Mar. 8, 1950	A	Warm	High	20	25.0±1	10	50	100
	to April 19, 1950	B	Warm	Low	20	25.0±1	1	5	... .
		C	Cool	High	20	15.0	1	5	100
		D	Cool	Low	20	15.0	1	5	100

versely affected by the high temperature. He also reversed his experiments; that is, he placed oysters which had been held in high temperature water in cool aquaria, and vice versa. He obtained essentially the same results after this switch as were obtained in the original experiments.

Owen concluded that "low temperature stops or retards the pathological action of the parasite." He believed that high temperature was an "absolute requirement" before *D. marinum* could kill oysters. This is not quite true, for many deaths from *D. marinum* have been recorded in cool weather, but the incidence of deaths due to *D. marinum* in cool weather is certainly very low. This was demonstrated by Mackin and Sparks (this volume, last paper, 1962). At seven stations in Barataria Bay, Lake Grande Ecaille, and Billet Bay, each containing a population of 1000 oysters, death rates due to *D. marinum* were shown to increase from a mean of five oysters per 1000 population in March, to a mean of 207 per 1000 population in August. Mean water temperature at the Grand Isle Laboratory in March 1957 (the year of the study) was 17.6°C while in August it was 29.2°C. Bay water temperature possibly varied ± 1°C from the temperature in Bayou Rigaud, where the readings were taken. It is apparent that under tray conditions in natural waters the effect of temperature is much more marked than it is in aquaria, for the August death rates were 41 times higher than March rates. In outdoor trays there was more reduction of the death rate in cool water; the warm period death rates were as high in the field trays as in the laboratory aquaria. The time required to reach high levels of mortality was several times as great in the field as in aquaria.

In Chesapeake Bay, the effect of winter cold is much more marked than it is in Louisiana (Andrews and Hewatt, 1957). The disease practically disappears in winter. Temperatures in the lower Chesapeake Bay in winter (Hewatt and Andrews, 1954b) range from 3° to 8°C in January and February and do not rise above about 12° to 15°C at any time in March. The peak summer temperature in the Chesapeake area is about 30°C. but the duration of high summer temperature (less than 2 months) is much less than in Louisiana (about 6 months).

Andrews and Hewatt (1957) believed that *D. marinum* was "responding sharply to a temperature change from 28° to 30°C because greater numbers of gapers seemed to be retrieved following intensely hot periods." The continuance of high temperatures to the middle of October in 1954 seemed to them to cause excessive losses of oysters. Conversely, fungus-caused mortality slows noticeably when the temperature drops below 25°C and comes nearly to a standstill at temperatures below 20°C. Ray (1954b) commented on the abrupt cessation of mortality in a population of oysters kept at the Grand Isle Laboratory in the summer of 1953. From July 1 to October 27, 60 percent of the oysters died. Thereafter through most of the winter only negligible mortality occurred. The records of the laboratory show that on October 27, 1953, the first important cold front of the coming winter arrived, driving the water temperature down from 24°C to 17.5°C in two days. The sudden cessation of mortality with the first cold weather of fall has been observed several times. It is believed that 20°C is approximately the temperature level at which reproduction of *D. marinum* in the oyster host slows almost to a stop. The intensity of infection and the incidence in the host are little affected by the cold until mid- or late winter (in Louisiana) and may not drop measurably at any time during the winter if there are unseasonable warm periods such as occurred in Louisiana in 1956–1957.

Hewatt and Andrews (1956) studied experimentally the effect of high and low

temperatures on infection of oysters. Oysters held in cold water vats at 15°C were compared with oysters suspended in the York River in trays, where the water temperature was 26° to 28°C, and with others held in vats at 28°C. All oysters were artificially infected by feeding them tissue mince of heavily infected oysters. The study ran for about six weeks. The results of these studies are shown in Table 23.

The results of these studies confirmed findings of the author and Owen (1955), that low temperature drastically curtailed mortality rates, and also showed that infection itself was inhibited by the low temperature.

Andrews and Hewatt (1957) froze *D. marinum* in oysters but failed to kill the parasite after several days. The author froze *D. marinum* and found that numbers of viable cells decreased rapidly from the first day to the eighth, when all *D. marinum* were found to be dead as shown by failure to respond to thioglycollate culture tests. Near-freezing temperature for extended periods must certainly be lethal to the parasite or its range would extend farther north. It is believed that the absence of *D. marinum* from the Virginia seaside is due to exposure to successive freezes in winter. Oysters of the seaside are intertidal and the reefs are regularly frozen when exposed in winter. It is not known whether the oysters themselves are frozen, but they were observed by the author to be caked in ice several times in the winter of 1944–1945.

Salinity and Flushing Rates. Mackin (1956) reviewed data bearing on the relation of salinity to development of *D. marinum*. In that paper it was indicated that in Louisiana estuarine areas of mean low salinity of about nine parts per thousand, the fungus could exist in low incidence, but it is doubtful that epizootics develop in such areas. Below about nine parts per thousand, the fungus has not been found to exist. It was originally believed that the fungus developed epizootics only in relatively high salinity, that is, only above about 15 ppt. It is now known that *D. marinum* may flourish in mean salinity of about 12 to 15 ppt and probably lower.

It was suggested by Mackin (1956) that salinity may not be as effective in reducing incidence of *D. marinum* as is rapid flushing of an estuary. Evidence tending to substantiate that hypothesis continues to accumulate. But other evidence continues to confirm the concept that low salinity *per se* is effective in control also.

Mackin and Boswell (1953) investigated the effect of varying salinity on infection rate in oysters. Part of this study was carried out in the field, part in the laboratory. In the field study, oysters from Chene Fleur Bay were placed in trays, and one-half (200) were retained on the racks at the low salinity Chene Fleur field station and a similar number were held in trays at the high salinity field station, at the Texas

TABLE 23

Effects of temperature on infection and development of disease. Data from Hewatt and Andrews, 1956

Source of oysters	Temp. °C	No. of oysters used	No. dead	Percent dead	No. of dead infected	Percent of dead infected	Weighted incidence among dead
James River*	15	50	5	10	5	100	1.4
Rappahannock†	15	50	5	10	4	80	2.0
James River*	28	50	50	100	50	100	4.56
Rappahannock†	28	50	50	100	49	98	2.86
James River*	26–28	50	37	74	37	100	4.89
Rappahannock†	26–28	50	16	32	16	100	5.00

* James River oysters were from a locality where *D. marinum* does not occur.
† Rappahannock River oysters were from a locality where *D. marinum* normally is present.

A & M Research Laboratory in Bayou Rigaud. Live oysters from each station were withdrawn monthly to determine incidence and weighted incidence of infection of the populations, and careful checks of mortality rates were made for comparison with weighted incidence data. A sample of 25 oysters was sectioned at the start of the study to provide data on incidence and weighted incidence at that time.

Similarly, 100 oysters from Bayou Rigaud and Bassa Bassa Bay were held in trays at the Bayou Rigaud and Chene Fleur stations. These populations were also sampled on a monthly basis. In addition to the samplings of live oysters, gapers were recovered at the Bayou Rigaud station, and these were also sectioned.

The data showed little difference in infection rates in oysters from Chene Fleur Bay, Bassa Bassa Bay, and Bayou Rigaud. For this reason the data from all groups were pooled in the final analysis. The data from this study are summarized in Fig. 28. The Chene Fleur station has a mean salinity of 10 to 12 ppt with a range from 0 to 28 ppt; at Bayou Rigaud the mean is from 18 to 20 ppt with a range from 5 to 33 ppt. It can be seen from Fig. 28 that there is a very great difference in weighted incidence of *D. marinum* at the two stations. It is doubtful that all of this difference is attributable to salinity. Some part may have been due to difference in flushing rates, or to unknown factors. The difference between the winter and early spring months as compared with the summer months at the Bayou Rigaud Station is also clearly shown, but influence of temperature at the Chene Fleur Station is not apparent. Mortality due to fungus disease was very high at the Bayou Rigaud Station from June to September, several times as high as deaths from all causes at the Chene Fleur Station.

Two laboratory studies by Mackin and Boswell (1953) were run in the period June 27, 1951, to August 24, 1951. Fifteen oysters were kept in each of two aquaria supplied with running water at the rate of about 650 cc per minute. In the experimental aquarium, rain water and artificially desalinated water was used to reduce salinity to a mean of 11.1 ppt with a range of 8.6 to 14.8 ppt. In the control aquarium, only water from Bayou Rigaud was used. The salinity mean was 20.9 ppt and the range 17.6 to 25.0 ppt. Oysters used were 87 to 90 percent infected as shown by two preliminary checks.

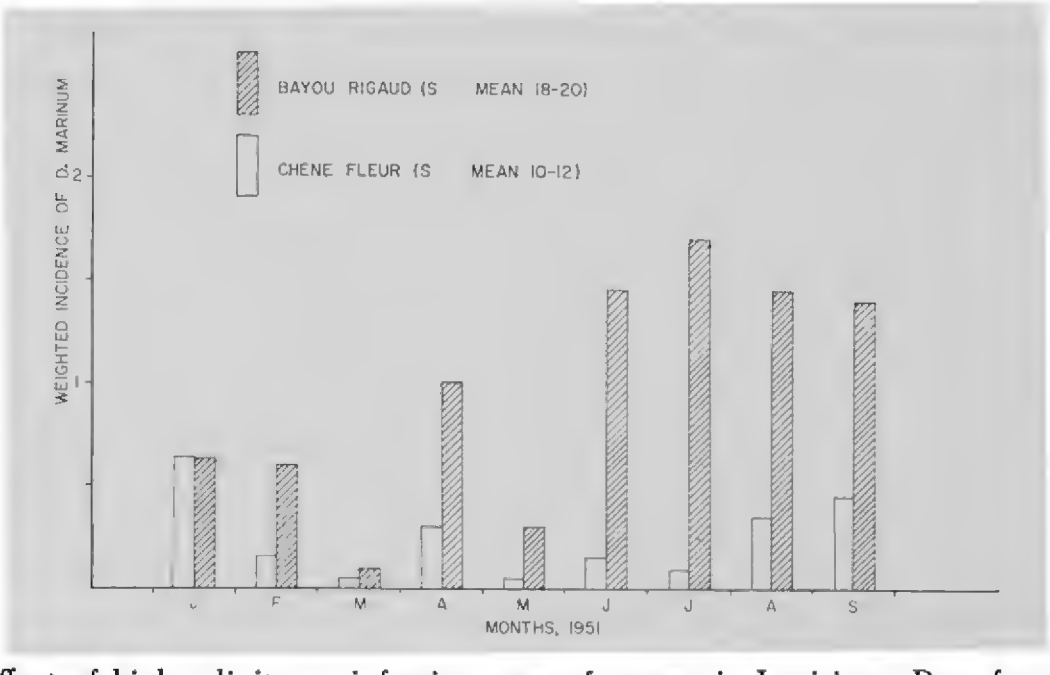

FIG. 28. Effect of high salinity on infection rates of oysters in Louisiana. Data from Mackin and Boswell (1953). Oysters were held in trays at Bay Chene Fleur (low salinity area) and compared with oysters in trays in Bayou Rigaud, Louisiana (high salinity area). Four hundred oysters were kept at each station and monthly samples were taken to determine the incidence and weighted incidence of infection.

A repetitive study, running from September 6, 1951, to October 23, 1951, differed only in that the salinity in the experimental aquarium was a little higher, range 13.6 to 20.8 ppt with a mean of 17.2. In the control salinity ranged from 19.8 to 28.1 ppt with a mean of 23.7 ppt. Data from these two studies are summarized in Table 24.

TABLE 24

Effect of lowered salinity in control of death rate and weighted incidence of *D. marinum*. Oysters used were from a population with incidence of about 85 to 90%.
Data from Mackin and Boswell, 1953

Aquarium no.	Salinity mean	No. of oysters	No. dead	Percent dead	Weighted incidence, gapers and survivors
1	11.1	15	15	100	4.40
2	17.2	15	5	33.3	3.20
3	20.9	15	11	73.3	3.47
4	23.7	15	8	53.3	3.60

These results failed to show an inhibiting effect of low salinity on death rate of oysters with high incidence of disease. The data are contradictory in that the two low salinity aquaria had radically differing death rates, suggesting the possibility that there was an inadvertent introduction of more oysters in advanced stages of disease in aquarium number 1.

Ray (1954b) studied the effect of low salinity in inhibiting infection in uninfected oysters. He used aerated aquaria adjusted to known salinity and infected his oysters by feeding them minces of heavily infected gapers. His data are summarized in Table 25.

TABLE 25

Effect of lowered salinity on infection rates of previously uninfected oysters. All studies were carried on in the summer of 1953 by Ray and Mackin, at the Texas A&M Research Foundation Laboratory at Grand Isle. Partially reported by Ray (1954b) and Ray and Chandler (1955)

Aquarium no.	No. of oysters used	Mean salinity ppt	Dosages of infective cells	Days to first death from *D. marinum*	Days to last death by *D. marinum*	Total dead	Percent dead	Weighted incidence
1	10	12.0	Very heavy	26	30	10	100.0	5.00
2	18	29.5	Very heavy	13	35	17	94.4	5.00
3	10	11.8	Moderate	52	90	10	100.0	5.00
4	10	11.4	Moderate	39	73	10	100.0	4.80
5	10	26.4	Moderate	37	62	10	100.0	4.05
6	10	11.9	Light	64	90	10	100.0	4.50
7	10	26.6	Light	38	76	10	100.0	4.20

The data again were contradictory; they seem to indicate that there may be a slight inhibitory effect of low salinity on development of *D. marinum*, but lack of consistency of the data causes doubt that there was even a slight effect.

Andrews and Hewatt (1957) pointed to the fact that in the Chesapeake *D. marinum* is usually absent from areas of salinity below 15 ppt. They reported that in the James River *D. marinum* was rare in an area of 17 ppt average in summer, but at Bell Rock in the York where salinity averages 14 ppt the fungus was moderately abundant. They moved heavily infected oysters from the York River to an area of the James River where the salinity range was 1 to 13 ppt. Controls were left in the York River, and disease-free oysters in the James River were used for comparison with the diseased oysters. This

treatment radically reduced death rate and weighted incidence in oysters moved to low salinity in May (when presumably the infection rate was low to start with), compared with the controls remaining in the York River. If the move to low salinity was made in August (when infection rate was high), no significant effect was observed. The authors concluded that if infection is once established it can persist in low salinity for a long time. The low salinity failed to reduce mortality in a heavily infected population from August 30 to November 5, 1954, although an uninfected population at the same station had a negligible death rate. The authors believed that low salinity itself is not an effective limiting factor on infection with *D. marinum.*

Although no specific experimental data are at hand it is thought that dilution by inflowing fresh water and rapid flushing rate are the factors principally responsible for the field correlation of low salinity and low incidence of fungus disease. The data presented by Andrews and Hewatt on incidences in the James River are particularly suggestive. It has been observed that concentrations of disease in Louisiana decrease rapidly in bayous subject to fresh-water flushing, while large bays with slow flushing rates have a more or less constant level of disease. Barataria Bay, with a flushing rate of about 21 days, is a good example of the latter case. It has also been noted that in late summer and early fall, severe epidemics may occur in bayous which normally have fairly low salinity. These epizootics coincide with the maximum tidal level, which rises from mid-winter to early fall. Influxes of large quantities of high salinity water occur in late September when local rainfall is diminishing in amount, and the more inland bayous often may be flushed in reverse, the net movement of water being inland rather than seaward.

Dawson (1955) found little evidence of a salinity effect on *D. marinum* incidence at 19 stations in Apalachicola Bay. Although Dawson's salinity data were too incomplete for proper analysis, it is pointed out that Apalachicola Bay has been reported by Ingle and Dawson (1953) to attain salinity in excess of 40 ppt. The excess over seawater salinity is due to evaporation which indicates that this bay has a very slow flushing rate. Dawson found *D. marinum* present at 17 of 19 stations checked, and a weighted incidence in some samples comparable to that found by the author in Barataria Bay.

Effects of Transportation and Planting. As far back as 1899, Moore pointed out that Louisiana oystermen plant excessively large quantities of seed oysters on their bedding grounds. According to Moore, plantings of 600 sacks (900 standard bushels) per acre were the rule. This practice is generally followed today. Sometimes even more than 600 sacks per acre are planted. This results in overcrowded beds. It is thought that the severity of epizootics is greatly increased by the density of the plantings. Seed oysters are largely susceptibles; when they are introduced to such areas as Barataria Bay where the disease is enzootic under optimum conditions the effect is often catastrophic. Year after year, great numbers of these susceptibles are introduced as seed into Barataria Bay and similar areas, tending to maintain disease incidence and intensity at a high level.

Zacharie (1898) first referred to losses of oysters in transportation. He stated that shock (including "heavy peals of thunder") was sufficient to kill oysters on boat decks. His statement that transportation of oysters to market in warm weather sometimes resulted in loss of entire cargoes has more possibilities of a correct cause and effect relation. Prytherch (in Galtsoff *et al.*, 1935) was more specific about losses of oysters in transportation. He stated that oysters in Terrebonne Parish which had been held on

holding grounds in the winter of 1932–33 gaped during shipment to market. Many gaped, according to Prytherch, while still on the holding grounds, their deaths coinciding with an unseasonably warm dry winter. McConnell (1933) fixed the total loss of oysters in Terrebonne and Lafourche parishes in 1932–33 at about 300,000 barrels. While there are no data to connect *D. marinum* to the losses of oysters in the early part of the decade, 1930 to 1935, it is suggestive that Owen (1955) established that *D. marinum* was present in about that period.

Several of the later holding-ground and transportation losses have been observed. One of these occurred in Creole Bay (a western sub-bay of Barataria proper). Oysters held on a "holding ground" died in large numbers when loaded for market (in October, 1949). The period was unseasonably warm. The load was refused by the New Orleans dealer to whom they were taken. They were returned to the beds in Creole Bay, but none survived the prolonged exposure. However, several oysters which had escaped the loading were taken by the author from the holding ground. Some of these gaped after a short time out of water, and all were found to be heavily infected with *D. marinum*. Similar cases have been observed in Lake Felicity, Barataria Bay, and Billet Bay.

It has appeared likely that the stimulus to this type of loss lies in the "holding" operation. Oystermen may harvest oysters for five to ten days at a time and "plant" them as they are culled on shell-hardened bottom near their oyster camps where they can be watched. These oysters are placed on the bottom in deep layers as much as 6 to 18 inches thick. In cool weather, this process appears not to be too harmful. But if the weather becomes warm, trouble often results. It is assumed that fall marketing may take oysters which have passed through a summer epizootic of fungus disease, and still have a high residual infection. This is latent as long as the weather remains cold. The combination of stacking in layers on the bottom, and warm weather triggers an epizootic which culminates often in excessive deaths after the oysters are loaded on boats for marketing.

In early November, 1957, oysters were stacked four layers deep in a running-water aquarium at the Grand Isle Laboratory. In an unseasonable warm spell in the middle of November in which the water temperature reached 30°C, an epizootic of *D. marinum* occurred. Thereafter few deaths occurred until early March, 1958, when, with water temperature between 15° and 20°C, deaths from fungus disease mounted rapidly to a high level. A control aquarium (with only one layer of oysters) had significantly less mortality than did the "stacked" oysters and those experimental oysters in the bottom row more than doubled the mortality in controls (49% and 23% respectively). However, about 25 percent of the oysters dying in the experimental aquarium died of causes other than *D. marinum* infection. These were mostly in the bottom row. All oysters dying in the control aquarium died of *D. marinum* infection.

This experiment was in some respects not a valid duplication of field conditions. For example, the daily examination required that oysters be removed from the aquarium. This brought daily relief from the stacking pressure in the experimental aquarium. Gapers were removed daily, thus removing a source of infection. This probably was somewhat balanced against the detrimental effect of the confining aquarium.

Transportation effects were also tested in another way. Since the principal effect of transportation is holding out of water, this effect was tested by removing oysters from the water and checking for increase in intensity of infection at zero time and at measured intervals thereafter (Mackin, 1954b). Three separate studies were made. The results

of these are summarized in Fig. 29. In these experiments there were four control groups of 25 oysters each. These were checked for weighted incidence at zero period. The mean weighted incidence in these four control samples was 1.62 and the range was from 1.38 to 1.80. Out of 13 experimental groups of 25 oysters each, only one (held out of water for 12 hours) fell within the range of weighted incidence of the four control groups and it was on the upper end of the range (weighted incidence of 1.80). All other samples held out of water exceeded the upper limit of the range in controls by significant amounts, and there was a general increase in weighted incidence from the twelfth to the eighty-fourth hour. These studies were all carried out in cool weather, beginning on March 4, 1953, and ending on May 9, 1953. Oysters were kept in the laboratory and shaded from the sun. It is believed that holding out of water in hot weather would greatly accelerate the increase in disease intensity, accelerating the rate of increase in weighted incidence. In Louisiana, seed planting begins on September 1, when disease incidence is maximum, and while the weather is still hot. So far as known, the effect of transportation under actual field conditions has not been tested, but it seems probable that *Dermocystidium* infections are intensified as they were in these experiments, and often more rapidly.

D. marinum AND THE OYSTER INDUSTRY

Within the range of *D. marinum*, and especially in those states where extensive planting of private leases makes up most of the industry, disease caused by *D. marinum* is perhaps the principal cause of losses of oysters. Virginia and Louisiana are the two states where researches have established that the fungus is of paramount importance. In both states it is desirable that new methods be worked out which will utilize known

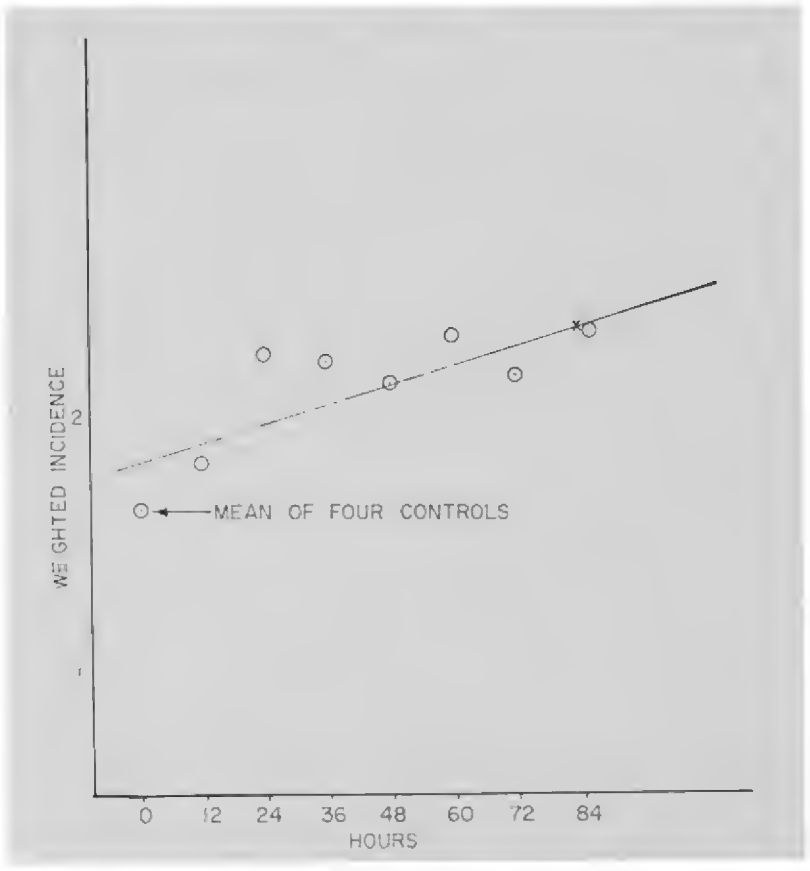

FIG. 29. Effect of holding oysters out of water on weighted incidence of infection. The curve summarizes the results of three repetitive experiments in which oysters were held out of water and the weighted incidence measured in samples at intervals as indicated on the graph.

epizootiological factors in reducing the summer epizootics. Some progress has been made in that direction. Ray (1954) pointed out several methods by which it may be possible to effect some improvement. Better methods of oyster production are needed for the entire range of the parasite.

Certain established methods of combatting disease in commercial fisheries animals are known. Some of these show promise in the case of *D. marinum;* others appear now to have little or no practical value. Basic methods in combatting disease are as follows:

Treatment of Infected Populations with Specific Fungicides. This method appears to have the least merit in the light of present knowledge and present oyster culture practices. It is difficult to conceive of a fungicide so selective that it could (a) do no harm to the host oyster, (b) produce no significant changes in the community of organisms associated with oysters and especially those which are themselves valuable for sporting or commercial reasons, and (c) cause no reduction in food supply. Other obvious problems are those of costs of application, and the large amounts needed to maintain effective dosage in the face of enormous dilution. Nevertheless, there may come a time when the present wasteful hit-or-miss methods of planting and harvesting may be impossible to maintain. The European *O. edulis* industry has already reached that point. As methods of oyster production grow more refined, as they are in Holland and France, opportunity for effective treatment by specific fungicides may appear. For example, marketing of oysters in Holland is now done from reserves stored in artificial holding basins, which would be ideal for concentrated treatment. There are more and more attempts in our own country to produce oysters in ponds of limited size (Lunz, 1946) where conditions may be controlled. It is conceivable. therefore, that in time the oyster industry will have developed to a stage where fungicide treatment will be economically feasible. It would seem desirable, therefore, to take the first step looking toward that end. That would be to sift the large number of compounds available, and to select those found effective as fungistatic or fungicidal. If such compounds are found and are available in quantity at low cost, the second step would be to explore the possibilities of developing methods of application to large numbers of oysters in a retaining basin of some sort. Oysters can stand temporary storage of very large numbers in small areas, and under very adverse conditions. This fact may be the key to successful treatment.

Management of Oyster Culture so that Ecological Conditions Are Optimum for Oysters and Minimum for the Parasite. The best available way to take advantage of good conditions for survival and rapid growth is to shift oysters from place to place. In Louisiana the best sets occur where predation and disease are worst. To utilize these high-setting areas, provision must be made to move young oysters immediately after setting to low salinity areas. Heavy sets occur in lower Barataria each year, but the young oysters are destroyed in the first few weeks by conchs. These young oysters would become an asset to the industry if moved within a few days after setting. Dependence on natural seed reefs which must be State-maintained would not be necessary if the natural set on "barren" reefs in high salinity areas could be used. This would involve development of lattice-work barges, floated by air-filled chambers, in which culch could be systematically piled. The moving of a train of such barges to low salinity areas by an oyster lugger would entail small expense. Following a move the young oysters would have to be maintained in low salinity waters until the summer high temperature periods were past. At the end of the summer they should be returned to high salinity waters

where growth during the winter months is more rapid. This first winter probably should be passed through in trays mounted on racks (Anonymous, 1940). This would facilitate removal to low salinity in early spring, just prior to reaching one year of age.

These oysters in Louisiana waters would now be two inches to three inches long, single and far better stock than the usual seed oysters from the natural reefs. They would be of uniform age, uncluttered by heavy set of young oysters, and with less fouling than usual. But they must be returned to low salinity water prior to (a) the oyster setting period, (b) the advent of the new crop of young conchs, and (c) the beginning of fungus epizootics. If in trays, transport in the lattice-work barges would be easy. In the fresh water they should be again placed on racks in trays. Toward the end of the summer, they must be returned to high salinity waters and planted on the bottom. It is at about this time that trays will become so overcrowded that it would be necessary to redistribute them anyway. By the middle of the winter these oysters will be ready for market as raw shucked stock.

Repeated removal of large numbers of oysters is expensive and the economics of the plan are uncertain, but oysters moved by means of special equipment in trays may be less costly than dredging them off the bottom each time they are moved. If the loss to fungus disease is reduced by one-half, the total crop will be more than doubled. It may be that increased yield will more than offset the increased costs. The increased value may be more than the value of the oysters saved. It will also increase as the quality of oysters is increased.

Cauthron and Mackin (1953) studied methods of production in Barataria Bay, assuming that the process of moving oysters would be too expensive for general use. These authors found that when oysters set in the late summer (last part of September or early October), the young oysters would pass through the following summer without undue loss to fungus disease because they would be only 8 to 10 months old. These oysters were marketable prior to the second summer when the very heavy losses from fungus disease normally occur. Spat for such culture must be taken on artificial culch (egg-crate fillers primed with a light cement). They must also be held in trays through the first winter, but may be planted on the bottom when one year old which would be in the late summer or fall. This method is successful in some fast-flushing bayous, where salinity is high but where conchs are not numerous because of the flushing action, including more than one-half of all the Louisiana oyster-producing area. This method also has the advantage that dependence on the natural seed reefs is eliminated, but it has the disadvantage that tray culture is necessary.

Selection of Naturally Resistant Strains for Seed Stock. The discovery by Andrews and Hewatt (1957) that oysters from South Carolina were resistant to effects of fungus disease to some extent provides some hope for development of resistant stock, locally or by regular importation of resistant seed from South Carolina or elsewhere. There are some data that indicate that populations exposed to disease in enzootic areas are more resistant to infection and lethal disease than are stocks from non-enzootic areas. The principal stumbling block for development of *in situ* resistant stocks is that importations of susceptibles must be strictly banned. There is also the problem of preventing natural hybridization of resistant strains with susceptible stock from low salinity areas. The larvae of these susceptibles may be carried long distances. This one factor may make the development of resistant stocks in some areas impossible.

Regular introduction of resistant seed probably has greater possibilities than has the

development of resistant stocks. If it is true that South Carolina seed are resistant to some degree a realistic approach to the problem is available.

Objections to the *in situ* development of resistant stocks may largely disappear when and if pond cultures of oysters on a large scale become a reality. Pond culture itself may be the only medium through which sufficient control of cross-breeding can be exercised to make possible the in-breeding of resistance to disease. The steps in development of resistant strains must be in this order: (a) selection of resistant individuals from natural populations, (b) cross-breeding in aquaria, where all factors can be controlled, (c) transfer of the product to closed ponds for development of large volumes of resistant stock, and (d) transfer to field conditions. As in other methods of disease control, a high degree of artificiality is required, along with increased cost of production.

Other Parasites and Diseases

Studies on *Nematopsis*

Prytherch (1938b) published an account of the life cycle of *Nematopsis sp.* which he found parasitizing oysters, and later (1940) described this gregarine as *Nematopsis ostrearum*. Prytherch believed that "heavy infections were found with serious oyster mortalities". These mortalities he indicated were in Mobjack Bay, Virginia, and Lake Barre, Louisiana. He claimed that high concentrations of spores were found in weak and dying oysters. These, when in adductor muscle, gill and mantle, were accompanied by paralysis, gaping, and stunting of shell growth.

Prytherch (1938a, 1940) performed experiments testing lethality of *Nematopsis* in aquaria. He used many experimental oysters and exposed them to infection by placing crabs in the aquarium with the oysters, or in an adjacent aquarium. Control oysters were not subjected to infected crabs. The results of six of Prytherch's experiments are tabulated in Table 26.

Prytherch (1938b) also performed duplicate experiments to these in the spring of 1938. These had lesser mortalities by 10 to 20 percent, but did not differ otherwise. Prytherch used oysters from very low salinity areas for all these infection experiments but got his infected oysters for infecting crabs from high salinity areas near Beaufort, North Carolina, a significant factor when considering his results. He fed infected oysters to his crabs in the experimental aquaria. These oysters were, therefore, placed in the

Table 26

Results of experiments by Prytherch (1938a, 1940) in which oysters were infected by *Nematopsis* sp. by exposing them to infected crabs. All studies lasted about three months at Beaufort, North Carolina, September to November, 1937

Experiment no.	No. oysters used	Control or experiment	Crab host used	Mortality, percent
1	250	Control	None	7
2	250	Experiment	*E. depressus**	67
3	250	Experiment	*P. herbstii*†	84
4	250	Control	None	3
5	250	Experiment	*E. depressus*	46
6	250	Experiment	*P. herbstii*	71

* *Eurypanopeus depressus*.
† *Panopeus herbstii*.

experimental tanks and were shredded by crabs. There were none in the controls. It seems almost certain, therefore, that he used a highly successful method of infecting his experimental oysters with both *Nematopsis* and *Dermocystidium marinum*. The latter is now known to parasitize oysters at Beaufort. Prytherch could not at that time recognize an infection method for a fungus disease which was not discovered until ten years later.

Sprague (1950) and Sprague and Orr (1953) described extensive experimentation with *Nematopsis*. Their studies were carried out at Grand Isle, Louisiana, in 1947 and 1948. They had considerable success in infecting oysters and the alternate crab hosts and discovered that *N. ostrearum* produced those infections which were found predominantly in the oyster mantle, while a new species, *N. prytherchi* Sprague (1949) was found to parasitize predominantly the gills. The crab hosts of *N. ostrearum* were *Eurypanopeus depressus*, *Eurytium limosum* and *Panopeus herbstii*. The crab host for *N. prytherchi* was found to be *Menippe mercenaria*.

In their studies of the effect of *Nematopsis* on the oyster host, Sprague (1950) and Sprague and Orr (1953) had results very much like those of Prytherch (1938), excepting that they had much greater mortalities in controls. In some cases, control mortalities exceeded those of the experimental aquaria. These authors were able to recognize that there was a mortality factor in their experimentation other than the possible effect of *Nematopsis*. They were also skeptical that mortalities resulting from *Nematopsis* occurred under field conditions since the infections produced artificially in the aquaria were incomparably greater than any found under normal conditions.

The results obtained by Prytherch, Sprague, and Sprague and Orr show that the laboratory experimentation did not give a certain answer to the question of whether or not *Nematopsis* was a factor in mortalities of oysters on planted beds in Louisiana. In no case was the experimentation free of the possibility that mortality factors other than *Nematopsis* were involved, as indicated above. Some other experimentation indicated that *Nematopsis*, with the infection intensities commonly found on oyster beds in Louisiana, is not a significant factor in mortality. Roberts (1948) compared the intensity of *Nematopsis* infections in a large number of gapers with the intensity in survivors. The oysters were kept in aquaria at the Grand Isle Laboratory. When an oyster died, *Nematopsis* spore counts were made in mantle, adductor muscle, and heart. Results were reported as the mean number of spores per low power microscope field, ten fields being counted. There were two studies, each using oysters of different origin. The intensity of infection in gapers was compared with the intensity of infection in random-selected live oysters from the same lot. Roberts' data are summarized in Table 27.

TABLE 27

Comparison of intensity of *Nematopsis* infection in gaping oysters with intensity in controls (live oysters)

			Spores per field, mean of the mean of 10 counts in each oyster					
			Mantle		Muscle		Heart	
Study no.	Live or gapers	No. of oysters checked	Total fields counted	No. per field	Total fields counted	No. per field	Total fields counted	No. per field
1	Live oysters	12	240	67.5	240	11.3	120	2.2
	Gapers	64	1280	55.5	1280	12.7	680	0.6
2	Live oysters	13	260	63.5	260	57.6	130	3.0
	Gapers	47	940	45.5	940	20.6	470	0.9

These data show that infections of oysters by *Nematopsis ostrearum* were no heavier in oysters that died than in live oysters. Since *N. prytherchi* infections are concentrated in gill tissue, this was not a valid test of this species.

Extensive sampling of oysters from various beds for numbers of *Nematopsis* spores by Mr. Dan Wray of the Research Foundation (1) failed to show really heavy concentrations anywhere, (2) showed light infections nearly everywhere, and (3) failed to show any correlation of intensity of infection with mortality rates in different areas.

Owen, Walters, and Bregan (1952) found all oysters examined in Louisiana to be infected with *Nematopsis ostrearum*. Oysters from low mortality areas east of the Mississippi River had somewhat heavier infections than those found in oysters in their "high mortality" area between the Mississippi River and Bayou Lafourche. They also found an increase in infection intensity in summer months at low mortality stations without correlating increase in mortality rates.

The author has examined several thousand slides (sections) of oyster tissues. Most of these sections show a few spores of *Nematopsis*, both *N. ostrearum* and *N. prytherchi*. In no case have significant histopathologies been observed in association with the spore cysts. No tissue reaction results from the spores, which do not reproduce in oyster tissues. There is no evidence that the spores produce toxins of any kind (as Prytherch believed). Concentrations such as those claimed by Prytherch (1940) have not been found in oysters, other than those experimentally infected. He thought that spores in heavily infected oysters reached concentrations between 1 and 5 millions. Simple calculations would indicate that in an oyster 5 cms long, the number of spores per 10-micron cross section would be 500 if there were 2½ million spores in the whole oyster. But only a maximum of a couple of dozen spores has been found in any one section, though thousands of sections have been studied.

"Mycelial" Disease

On June 27, 1950, the author collected oysters from the Fulton Beach area of Aransas Bay, Texas. According to Dr. Gordon Gunter, oysters on reefs off Fulton Beach had recently been dying at a rapid rate, and oystermen had made complaints. No dying oysters were found, but all oysters were very thin, some almost glassy. About a dozen of these oysters were fixed in Zenker's fluid and later sectioned and variously stained. In those stained with Delafield's haematoxylin, a peculiar mycelium-like growth was found, but the staining preparations were not good, and stains other than Delafield's failed to stain the organism at all. No further studies were made at that time.

In the several years following, the mycelium was observed several times in gaping oysters from experimental aquaria, but the observations were so isolated that effective study could not be made. In 1953 oysters in several aquaria died very suddenly. Diagnostic cultures for *Dermocystidium marinum* showed that this fungus did not cause the deaths. Survivors from some of the aquaria were isolated in an aquarium especially reserved for the purpose, and the oysters as they died were replaced by fresh oysters in an attempt to keep the unknown lethal agent passing from oyster to oyster, so that material would be available for study after the busy period. Unfortunately time did not permit sectioning and study of the gapers at the time, but they were fixed and preserved. Later some of the oysters which died in this aquarium were sectioned and stained with

Harris haematoxylin. These preparations showed that the mycelial organism had been carried in this aquarium of oysters for more than a year before it was finally supplanted by *D. marinum*.

In the period from March, 1957, to September, 1957, several field studies using trays of oysters were carried out in Barataria Bay and adjacent bays. Stations were checked for mortality weekly and a high percentage of gaping oysters was recovered for study. At one station (Sugar House Bend in lower Barataria Bay) the records showed that there was a period in May when a large number of oysters died, and the culture records showed that this mortality was not caused by *Dermocystidium marinum*. This population of oysters had a very low incidence of fungus disease until July, when the usual summer epizootic set in. A total of about 20 percent of the 1000 oysters at this field station died in May. The abrupt onset of mortalities is shown in Fig. 30A, which traces the spring

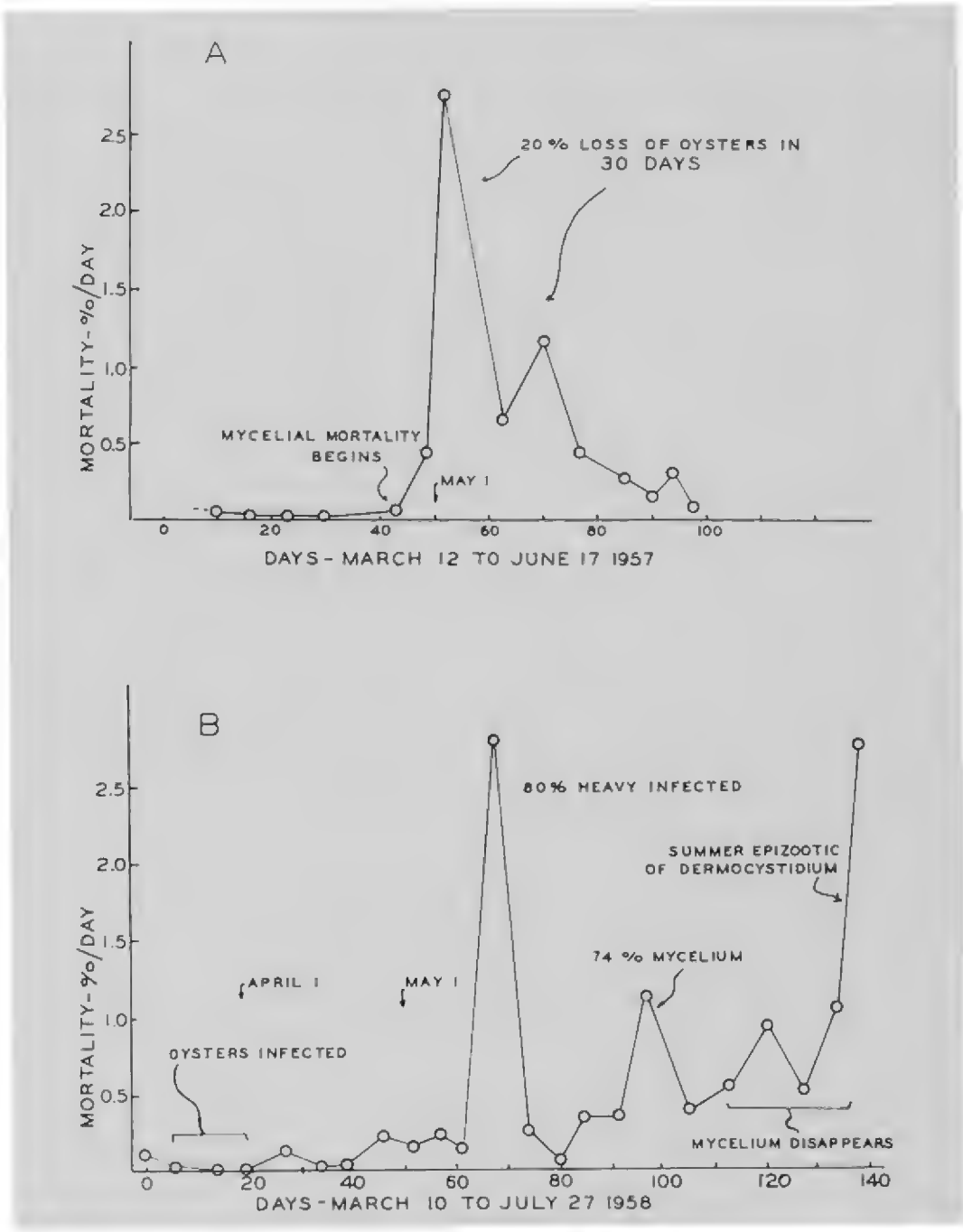

FIG. 30. A. Epizootic which may have been caused by "Mycelial disease" in Barataria Bay, Louisiana, 1957. B. Epizootic caused (?) by "Mycelial disease" in Bayou Rigaud, Louisiana in 1958. Compare with Fig. 30A.

mortalities by approximately weekly periods. This graph shows that immediately after the mortalities began, more than 10 percent of the oysters at the station died in a 10-day period. One hundred eight gapers were recovered (39 percent of those dying). The dead

and dying oysters were sectioned and the slides stained with Harris haematoxylin, Giemsa, and Heidenhain's iron haematoxylin. Sections were cut through the visceral mass just posterior to the base of the palps. About 75 percent of these oysters was found infected with the mycelial organism. It is believed that the incidence was actually higher, since the sectioning method is a rather small sampling of the entire oyster, and staining methods had not been worked out completely.

Initiation of Special Studies of the Mycelial Organism. The above-mentioned studies definitely associated mortalities of oysters with infection by the mycelial organism. However, except for the original dozen oysters from Aransas Bay, all oysters studied were gapers, that is, were either dying or already dead. The Aransas oysters were taken after the mortality was substantially over. It became desirable to determine definitely whether the mycelium attacked only dead and dying oysters, or was in fact a parasite, attacking healthy oysters, having a sequence of invasion and development in the host. To accomplish this end, several populations of oysters were placed at selected field stations in the Barataria area, one of them at the laboratory at Bayou Rigaud. About 800 oysters, including lots from several outlying areas, were placed in trays at each station. The stations were set up in January, 1958, and regular weekly checks were made beginning in early February and ending in early August. Samples of live oysters from each station were removed each week and fixed in Zenker's fluid for sectioning. All gapers collected were also fixed. The posterior gut region from each of these oysters was cultured in thioglycollate by the Ray method to permit separation of *Dermocystidium* disease from mycelial disease. The populations of oysters were selected on the basis of very low initial infection level with *Dermocystidium marinum*, to reduce the complications due to impact of two diseases on one population.

A typical explosive-type epizootic developed at only one of the 1958-period stations. Very conveniently, this one was at the laboratory in Bayou Rigaud. Only data from this station are presented here. Fig. 30B presents in graphic form the structure of the mortality developed at this station.

Development of Mycelium in Live Oysters. Previous studies had shown that a Harris haematoxylin stain was best to show the mycelia. A slight overstain, using more acid than usual, and a quick bluing with strong sodium carbonate was found to be selective and provided a clear picture of the parasite. This method was used in staining all oysters for diagnostic purposes.

Sections of live oysters showed that the attack of the mycelial organism began in early March. First infections were usually on the external epithelium of mantle, palps, or gills. Colonies of the mycelia were found on any external epithelial surface (Fig. 31A). They appeared to be loosely attached by a gummy material, and the filaments radiated outward, branching in a shrub-like manner. Frequent penetrations of the epithelium occurred. Some early infections were in the stomach or elsewhere in the gut. Nearly all infected oysters had internal as well as external lesions, but in the early spring these did not usually produce extensive infiltration in the tissues. Later, in April, the internal infections were more prominent, and live oysters were often found to have extensive growths, especially in the digestive gland.

Sixty live oysters were sectioned, which were taken in the period from March to June inclusive. Of these, the mycelium was found in 56, about 93 percent of those studied. It is believed that the incidence was actually 100 percent from inception of the attack

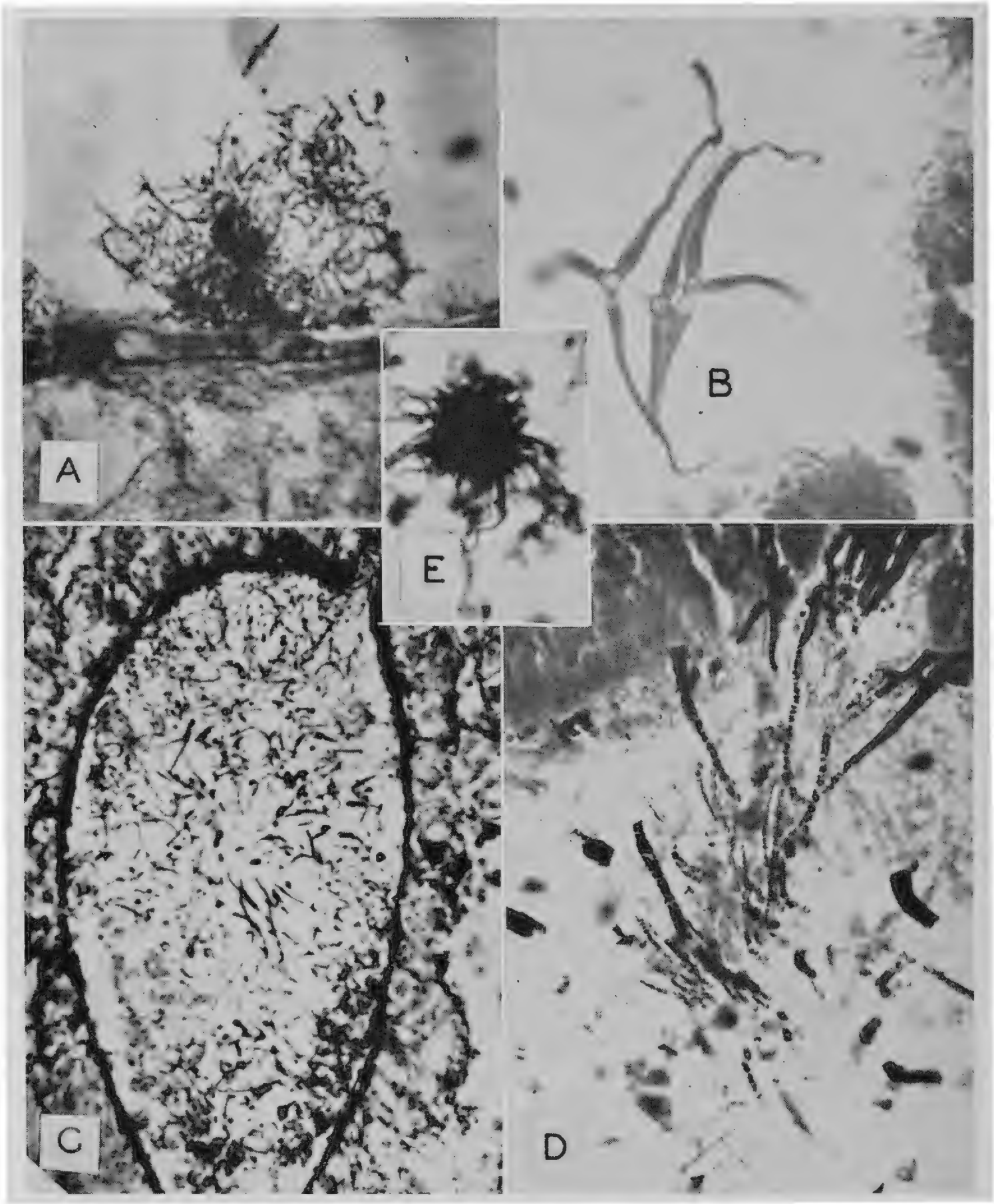

FIG. 31. Mycelial disease in oysters. 31A. A clump of mycelia growing attached to the external epithelium (mantle) of an oyster. Section stained with Harris Haematoxylin. 31B. Stellate bodies from mycelial disease in gut of an infected oyster. These structures vary in length (from tip to tip) from 5μ to 30μ, and may be up to 3 to 4μ thick near the base. 31C. Thick mass of mycelia in a blood vessel of a gaping oyster. Here the parasite resembles an Actinomycete. 31D. Fragmenting mycelia in tissues of an oyster. These probably represent degenerating parasites. 31E. A solid staining, rounded body with strands of radiating mycelia.

in March until late June. In July, incidence dropped sharply; more than 50 percent of the live oysters sectioned and stained were found to be free of the parasite.

Attack on oysters in March, when Louisiana oysters are in their best condition, established the mycelium as an aggressive parasite. Gonadal development is rapid in

that month, and shell growth also is good. The parasite disappeared in late summer, when the oysters are at their poorest.

Infections in Gapers. All of the 15 gapers recovered at the Bayou Rigaud station in February were found to be negative. Beginning with gapers collected on March 4 to the middle of July, all gapers but 2 (out of 70 sectioned) were found to be infected. After the middle of July only 7 out of 19 sectioned were found to be infected and *Dermocystidium marinum* infection was sometimes found mixed with the mycelium.

Again, the data strongly suggest that the entire population of oysters kept at the Bayou Rigaud station became infected in the early spring, and that all oysters carried infection to early summer. Involution forms of the parasite began to appear in June and July. Sections of oysters in the late fall and winter (November and December) showed that the mycelium had completely disappeared some time after the end of July.

Characteristics of the Mortalities. No experimental studies have been made to test the degree of pathogenicity of the mycelial organism. However, the circumstances are strongly suggestive that it is in fact the cause of the mortalities because, so far as can be determined from stained sections, it is the only associated organism which was shown to attack the live oysters and to develop a progressively greater intensity of attack on the host as the epidemic developed. On the other hand, certain bacteria have been found to be more or less constantly associated with the gapers, and their presence in live oysters has not been disproved. The hypothesis of cause and effect, therefore, is only tentative. But certain features of the associated mortalities are of such interest that they are inserted here without the implication that the etiology of the deaths is proved. The most interesting of these is the selective deaths of male oysters. Although random selection of live oysters from the population prior to the onset of heavy mortality showed that males and females were almost exactly equally represented, males made up 82 percent of all the mortalities in the period of April, May and June. Females made up 15 percent, and asexual oysters 3 percent. These latter were mostly parasitized by *Bucephalus cuculus* or *D. marinum*, both of which have a castrating effect. The heaviest mortalities in both 1957 and 1958 were coincident with spring spawning. The sections showed that gonads of gapers and live oysters alike were filled with eggs or sperm.

The very rapid and explosive nature of the mortalities at the peak period is without parallel. In 1957, 20 percent of the oyster population at the lower Barataria Bay Station 6 died in May, and most of these deaths were concentrated in the first 3 days of the month. In 1958 nearly 20 percent mortality occurred in a week and most of these died in a 3-day period. In the latter year, 104 gapers were removed in 3 days from 3 trays containing a total of 655 oysters, and many boxes were removed in the same period.

It is believed that the reason for the sudden onset of mortality lies in the fact that the muscular contractions which expel eggs and sperm force large numbers of sperm through lysed openings into the sinuses of the blood system. Masses of sperm were often found in the blood of dead males, and sometimes eggs were found in blood vessels of females. Protein shock and much mechanical damage could result from large numbers of active sperm in the blood. Distended walls of the blood vessels were often observed to be penetrated by the filaments of the mycelium.

Although the greater part of the deaths was associated with the spawning period, the data also showed a significant low-level mortality from March to July, associated with heavy infections of the mycelial organism. It is believed that mortality need not always be explosive.

Morphology of the Parasite, and Histopathological Effects on the Host. The parasite grows as a filamentous mycelium, branching irregularly in an arboreal fashion (Figs. 31A, C; 32, 33). When only the fine filaments with their branchings are seen, it looks and stains like an actinomycete. The natural color is a pale straw-yellow, and the cytoplasm has a refringent appearance when unstained. It stains lightly if at all with Heidenhain's haematoxylin and a number of bacterial stains, while coloring rapidly with acid haematoxylin stains, as in the Delafield or Harris method. The peculiar staining properties may have been caused by the Zenker's fixative, which was used with all oysters studied.

Associated with most mycelial growths are deep-staining amorphous masses (Fig. 32) which often appear to be part of the parasite, and may possibly be. Growths through such dense tissues as epithelia nearly always take place in the form of these blackened masses. These may be materials formed by tissue responses, which produce basic staining, react like the mycelia, and mask the mycelia themselves. Many small, yellowish, crystalline granules invariably are found associated with the parasite. These often are left in tissues to mark the site of a former lesion.

The mycelia themselves vary greatly in size. Some have a diameter of less than 0.5 microns and are at the borderline of resolution with the light microscope. Others may have a diameter up to 5 or 6 microns. At intervals along the mycelia structures appearing to be joints are occasionally found. These may be at branching points, or the filament may be sharply angled where the "joints" occur.

No structures interpreted as nuclei have been observed. The basophilic reaction of

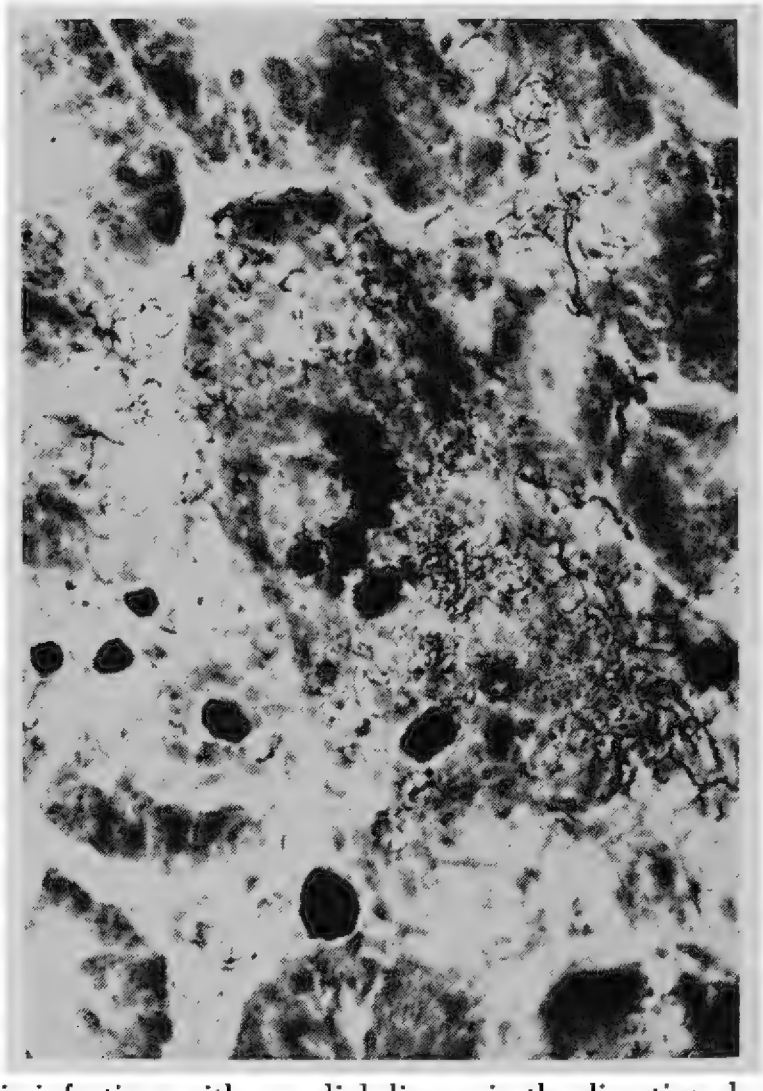

FIG. 32. Characteristic infections with mycelial disease in the digestive gland and Leidig cell tissue area. The irregular black bodies in the tissue are common signs of infection. The fine branching mycelia have nearly dissolved a branch of the digestive gland in the center of the photograph.

FIG. 33. Massed mycelia in the gut lumen of an infected oyster. The large mycelia measured 5μ thick at the thickest point. This photograph shows the peculiar type of branching sometimes observed.

the cytoplasm as a whole indicates a fine dispersal of chromatin elements. However, preparations stained with thionine may show a great many discrete, deeply-staining cytoplasmic granules of quite large size. The numbers, arrangement, and size of these granules do not suggest nuclei.

General appearance of the colonies in tissue is shown in Fig. 32. Lesions involving tubules of the digestive gland are shown. Fig. 32 is a photomicrograph of a section of a live oyster. In gapers, much more extensive networks are usually found, as shown in Fig. 31C, where a thick network of the parasites has occluded a blood vessel, or in Fig. 33, where masses of arboroid branches fill the lumen of the anterior loop of the gut.

In Fig. 31E, one of the deep-staining bodies appears to be branching in a radiating manner in transition to the mycelial form. Such masses are common and suggest that some of the branching structures may be jell-like and capable of retraction and projection like pseudopodia of the protozoa.

In many oysters "stellate" bodies (Fig. 31B) are found. These seem to be formed by germination in two or more directions from a single spore. The center from which the branches arise stain much more lightly than the branches themselves, as shown clearly in the photograph. Large "stellate" bodies were chosen for this photograph, but they occur in all sizes down to the very minute. Clear-centered spore-like bodies are often produced at the ends of very fine branches of the mycelium. It is assumed that the stellate bodies arise from these spores. The arms of the stellate bodies may often be found branching into typical arboroid colonies.

The spores referred to above are formed in a manner suggesting that found in the

genus *Micromonospora* of the Actinomycetes. They appear to be attached to small lateral branches.

In some slides, mycelia may be found which appear to be breaking into spore-like bodies (Fig. 31D). In this stage, the cytoplasm stains very densely, appearing black in sections. The "spores" have a pycnotic appearance, and it is believed that these represent dead portions of the parasites, or involution forms. They are useful for seeing the thin, clear, non-staining pellicle or wall of the filaments.

Growth of the parasite in the host produces little cellular reaction. Rarely are phagocytes aggregated at the site of lesions, and in no case has phagocytosis of parts of mycelia been observed. Those parasites found on external epithelia evidently lyse the underlying cells (Fig. 31A) causing them to thin out and completely disappear under a mass of parasites. Leidig cells in the vicinity of an actively growing colony take on a granular, crystalline appearance, and ragged break-down areas appear. Infected digestive gland acini simply disintegrate when occupied by a colony. Epithelia of the gonad, stomach, and gills likewise became granular in appearance and the cells dissociate. It is suspected that the parasite may be toxic to the host. Heavily infected oysters stop feeding, but those with only external parasites, or only scattered internal lesions seem not to be affected except locally.

Distribution and Epidemiology. Distribution in *Crassostrea virginica,* so far as now known, includes Texas, Louisiana, Virginia, Maryland and probably most states of the south Atlantic. It has also been observed in oysters, *Ostrea lurida, from* South Puget Sound, Washington. In Louisiana it appears to be increasing at present.

Very little is known about the environmental factors controlling the parasite. It appears to be rather definitely a spring form, but is not confined to spring months. Some heavily infected oysters have been found as late as August. Temperatures of the water in Louisiana bays from March to May, when the parasite blooms, range from 12° to 20°C in March, to 20° to 29°C in May. Summer temperatures ranges from 27° to 32°C (June to September). Only the coldest months seem to be completely free of the parasite.

As for the salinity, the only certain conclusion is that high salinities above 30 ppt are not inhibitory. In 1957, salinities were generally lower than usual in the period of the spring epidemic in Barataria Bay, at the Sugar House Bend Station, and were probably below 15 ppt for the most part. It is probable that salinity is not a limiting factor in distribution.

So far as is known, bottom conditions play no part. The disease has been observed most often in tray oysters, raised several inches over the bottom. The disease occurred in Aransas Bay in oysters on a hard, raised, natural reef. In Barataria Bay, the 1957 epidemic was in an area of hard-packed sandy bottom, and the bottom in Bayou Rigaud is soft silt.

Relationships. The "mycelial organism" may be related to the Actinomycetes. There are, however, very definite differences between our form and the known Actinomycetes. First, no actinomycete has been reported which approaches the size of the oyster parasite, and the very large branching forms do not branch as do the Actinomycetes, although the smaller mycelia do. The attached colonies (on the external epithelia) resemble the Caulobacteriaceae rather than the Actinomycetes, especially in the gummy or waxy basal substance. The "stellate" forms and the joint-like structures have not been reported for Actinomycetes although "star-shaped aggregates" have been reported in the Myxobacteriales (Stanier, 1940).

The oyster parasite resembles the Actinomycetes in the form of branching of the finer filaments, in the affinity for acid haematoxylin stains, the production of "sulphur" granules (*i.e.*, the yellow crystalline materials), the refractive and slightly yellowish color of the cytoplasm, the radiating colony form and the production of spores on the tips of short fine branches, as in the Micromonospora. The differences mentioned above may be due to the fact that all our material has been studied in the host organism, while it is customary for the orthodox researcher to record only what he finds in isolated cultures. However, it is suggested that there are also characters that remind one of the blue-green algae. A degenerate blue-green might have most of the peculiar structures observed.

Literature. There are several references in the literature to organisms resembling Actinomycetes in oysters. One is by Orton (1924b) where he reported work by J. W. H. Eyre (see also Eyre, 1923). Eyre found "*Cladothrix dichotoma*" in sick oysters and called it a "false branching" form. *Cladothrix*, as pointed out by Skinner, Emmons and Tsuchiya (1947), was once used by some authors as a synonym for Actinomycetes. However, it is doubtful that Eyre used the word in this way for his reference to "false branching" indicates that he was assigning his *Cladothrix* to the iron bacteria where this genus belongs. However, Dollfus (1922) says that Eyre's *Cladothrix* was a *Nocardia*, which is an actinomycete. Orton (1924b), using Eyre's *Cladothrix*, was not able to produce adverse effect on oysters in experimentation.

Pettit (1921a, 1921b) found what he thought was a *Nocardia* in sections of *Ostrea edulis*. He believed that the great mortality of 1919 to 1925 in English oysters was caused by this organism. He called it *Nocardia matruchoti*. Dollfus (1921) thought that Pettit's actinomycete was a normal cell reticulum, and denied that it was a parasite at all.

According to Dollfus (1922) A. G. R. Foulerton isolated a species of *Streptothrix* from the "visceral sacs" (digestive gland?) of *O. edulis*.

The author has seen an undoubted actinomycete in an oyster (*Ostrea edulis*) from Holland. It is quite possible that all of the fragmentary reports listed above had to do with the same organism, and it may be the same as that reported here. However, the probability is that they are not the same. It would be difficult indeed for the European authors to have missed heavy infections like those reported here if they occurred in *O. edulis*.

Bacterial Parasite of the Gastric Shield of the Virginia Oyster

So far as can be determined there is no mention in the literature of bacteria attacking the gastric shield of oysters. However, a bacterial parasite exists with a very wide distribution on the Gulf and East coasts of the United States. It is very common in Louisiana, in the Barataria Bay area. It is estimated that probably more than 90 percent of mature oysters there are infected.

The parasite is commonly found on the outside surface of the gastric shield (*i.e.*, away from the stomach epithelium) in the area where the end of the style rests. In preparations made from sections through the anterior viscera, and stained with a tissue modification of the Giemsa technique, the bacteria show on low power examination as a dark blue band along the shield section, and penetrating the shield, sometimes to considerable depth (Fig. 34). In the Giemsa-stained preparations, the gastric shield itself stains a light pink or bluish color. In preparations stained with haematoxylin or other

FIG. 34. Section of the stomach of an oyster through the gastric shield, showing masses of bacteria growing in the surface of the shield. Stain was with Giemsa tissue stain modification.

"non-bacterial" stains, the presence of the bacteria can be detected easily by the lysed areas in the substance of the shield.

The bacteria are shown by high power examination to be streptobacilli which sometimes have quite long chains (Fig. 35). No attempt has been made to culture these interesting organisms. It is apparent that their growth is conditioned by the proteins of the gastric shield itself and may be influenced by the enzymatic substances of the style.

Nelson (1918) called the gastric shield "chrondroid-like" which indicates that the proteins may be of the same general nature as the gelatine of vertebrate cartilage. Yonge (1926) thought that the gastric shield was composed of fused cilia. Shaw and Battle (1957) apparently acecpt Nelson's hypothesis that the shield is formed of chrondrin, but suggest that it may have an origin analogous to that of the lining of the foregut of insects, in which case the gastric shield would be chitinoid. At present it is believed that the matter is not settled. If the gastric shield is chitinoid, the bacteria nourished by it may be related to the chitinoclastic bacteria, that attack chitins in bottom muds, or those parasitic in the exoskeleton of marine crustacea (for references see ZoBell, 1946). The bacteria of the oyster resemble chitinoclasts described by ZoBell in that they adhere tightly to the surface of the gastric shield, and penetrate it.

So far as the author is aware the only chitinivorous bacterium to cause disease in commercially valuable marine animals is that described by Hess (1937). This one attacks lobsters on the West Coast of North America and according to the author, destroys large numbers of them. This disease is called the "soft shell" disease. There must be other such diseases.

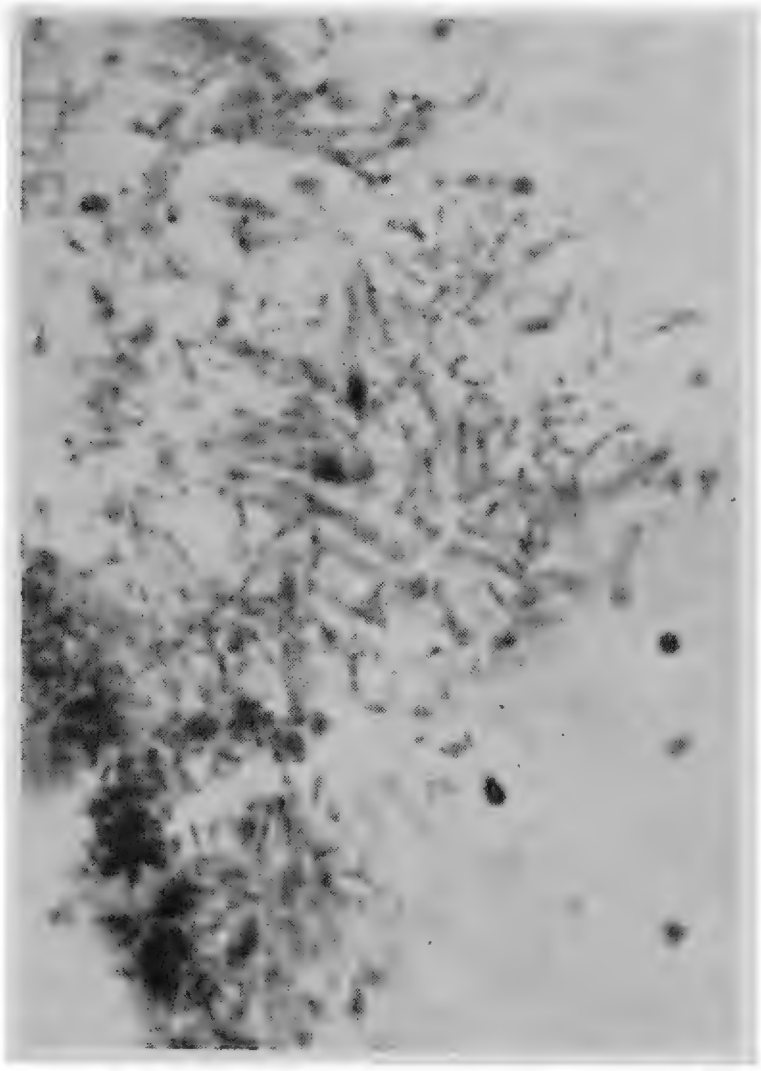

FIG. 35. High power field of a portion of the bacterial mass shown in Fig. 34, to show the chains of bacillae.

There is no reason to believe that this oyster parasite significantly damages its host. It seems to be well balanced as a parasite and occurs in oysters which are healthy and vigorously growing. However, the effect on the host has not been investigated. It is suggested that the organism would probably culture easily and might yield information of considerable basic value.

"AMBER DISEASE" OF OYSTERS

In 1956, two live oysters (*Crassostrea virginica*) which were observed to be a light amber color were fixed and sectioned. Slides revealed a parasite not hitherto seen in Louisiana oysters.

The two oysters were taken from a bed north of Lake Raccourci, in one of the numerous bayous draining into that lake. The parasite was not again seen until the winter of 1958–59. A small mortality of oysters took place in that period in Bayou Rigaud, and a number of gapers were taken which had the characteristic histopathologies of the original two oysters, and the same parasites. This type of oyster parasite now is of very considerable interest because of its possible relation to the "MSX" parasite being investigated in Deleware Bay and at other localities on the East coast. For this reason I am presenting a preliminary description of the parasite and the histopathologies associated with parasitization by it.

In Fig. 36A to D are shown various stages of the parasite as sketched from prepared slides. The most numerous stages are the more or less spindle-shaped amoebae. These commonly have from one to several filipodia or thread-like extensions of the cytoplasm,

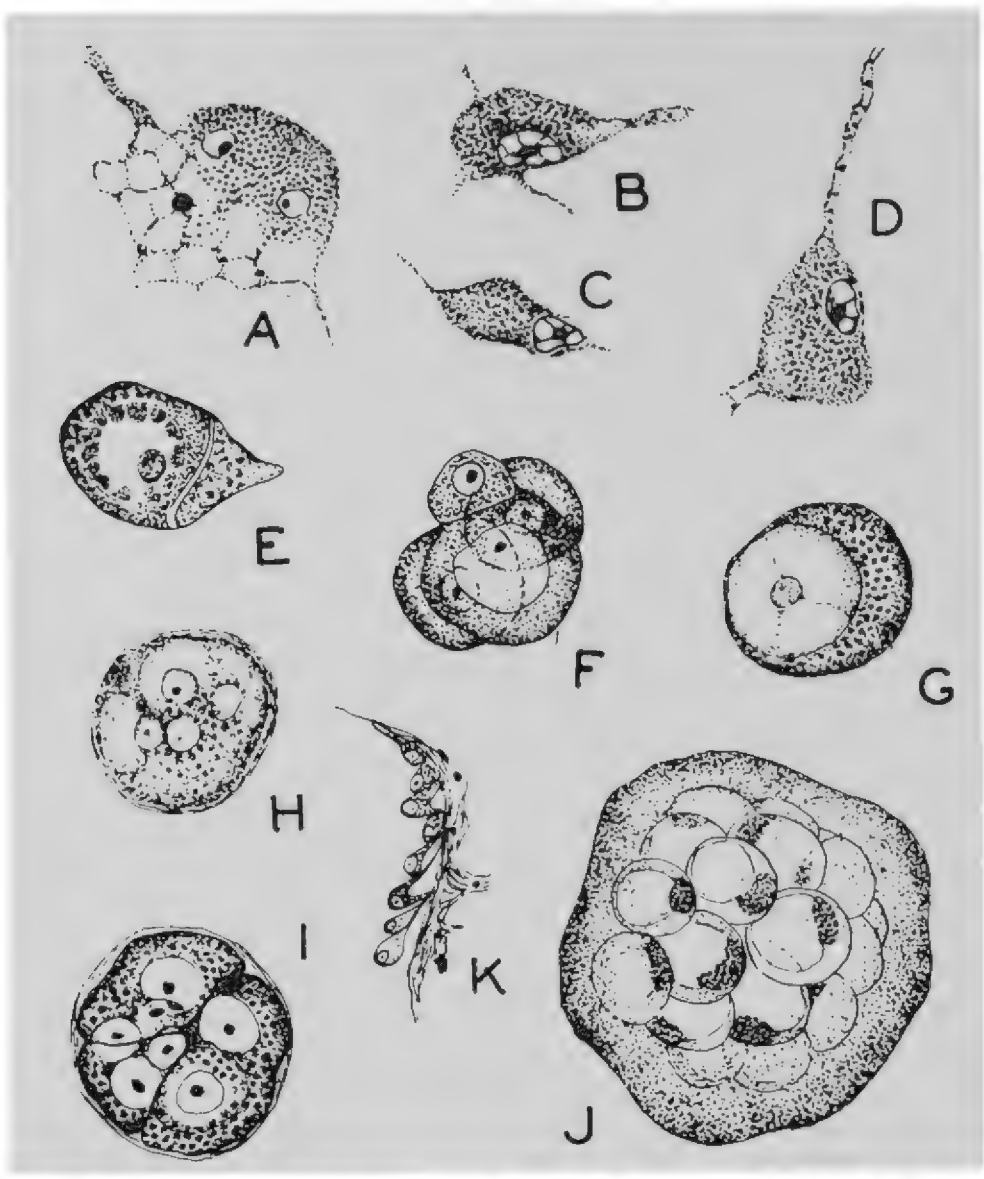

FIG. 36. Amoeboid organism causing "Amber disease" in oysters. 36A to D. Trophozoites as seen in oyster tissue, stained with Harris haematoxylin. The granules stain with any basic stain. 36 E, G. Enlargement of the nuclear structure prior to division in a trophozoite. The nature of the cap-like structure is not known. 36F. Aggregation, or result of division, in the amoeboid organism. 36 H,I,J. Various stages in nuclear multiplication and fragmentation. 36H. 4-nucleate stage. 36I. 8 (?)-nucleate stage. 36J. A cyst containing bodies that appear to be spores. 36K. Habit sketch of amoebae clinging to the wall of a blood vessel.

and one nucleus. Nuclei stain with difficulty with basic dyes and often appear to be vacuoles. However, a large number of fine granules in the cytoplasm are basophilic and may be stained with any ordinary nuclear dye. Accordingly the cytoplasm appears to be very dark, and the amoeboid stages are characteristically deeply stained.

The amoeboid stages are found mostly in the Leidig cell tissue, but may penetrate any tissue, and tend to congregate around the basal membranes of the gut and between tubules of the digestive gland. They often line the interior of blood vessels as shown in the habit sketch, Fig. 36K.

Reproductive stages are not numerous and are much less prominent than are the amoeboid stages. The reproductive process is initiated by nuclear enlargement, and some of these stages (Fig. 36E) can be seen to have a definite cap on one end. This cap may be explained in either one of two ways. It may be the cell remnant from which the migrating nucleus emerges just prior to sporangium formation, which is so characteristic of one group of the Synchytriaceae, or the cap may represent a fertilizing cell. In any event further enlargement of the nucleus takes place (Fig. 36G), followed by mitoses and nuclear fragmentation. In this early stage several nuclei will be found irregularly central in position (Fig. 36H). As the number increases (Fig. 36I) evident cleavages of the cytoplasm occur. Apparently these can end in fragmentation as shown in Fig. 36F,

or in the formation of a sporangium-like body containing spores as in Fig. 36J. These sporangia (?) are dark-staining, very thick walled, and irregularly rounded in form.

The histopathology is as striking as is the parasite. Most prominent are the masses of ceroid which form in the Leidig cell tissue. These granular masses stain dark green with Giemsa, black with Heidenhain's haematoxylin, and fail to stain (appear yellow) with acid haematoxylin. Destruction of the Leidig cell system is extensive, and there seems to be an intense cellular reaction. Liver tubules lose their normal pigment granules, shrink and become knobby-looking in cross-section. Infiltration of the various epithelia with parasites and leucocytes is common. Gonads atrophy, but since the parasites attack in mid-winter, it is not known how much of this to attribute to parasitism.

So far as now known, this parasite is found only in Louisiana. Outside of the observations given here little is known about pathogenicity, the capacity of the organism to produce epidemics, or other factors. It appears only relatively seldom, so far as is known; possibly it is in a marginal territory in Louisiana.

Watery Cysts of Oysters

A small percentage of the oysters in most samples taken in summer from high salinity areas have watery cysts in the visceral mass, palps, or mantle. These are not the same as the yellow pustules so commonly found in the adductor muscle or elsewhere, and have quite generally been overlooked in descriptions of diseased oysters.

The cysts, seen in the live oyster, are large conspicuous bubble-like cavities, generally distended by internal pressure of accumulated fluid. Sections show these cysts to contain a central granular material usually in concentric layers (Fig. 37A). Stained sections and smears from these watery cysts show a very small bacillus in the granular materials (Fig. 37B). A number of such smears have shown what appears to be the same bacillus, so it is believed that there may be only one causative agent involved. No effort has been made to culture this bacillus.

Recovery from the infection appears to take place through a sloughing of the cyst content. The cyst walls are formed of leucocytic cells which form themselves into an epithelium similar to that of the external epithelium of the mantle. When fully formed, the cyst ruptures, discharging the contents to the outside.

Yellow Pustule Disease

This disease is more or less frequent in oysters throughout the world, but there is no reason to believe that all the yellow pustules and abscessed areas found in oysters have a common etiology. The yellow pustules found in Louisiana oysters are much like those in the European "maladie du pied," and like the latter usually attack the adductor muscle.

Mackin and Cauthron (1952) found that in late summer 27 percent of oysters in the Barataria Bay area (five stations) had the yellow ulcerations and abscesses. More than half (56 percent) of the abscesses were found to be associated with penetrations of the shell by *Polydora, Martesia,* or *Cliona. Polydora* was most effective in producing abscesses, probably because it introduces mud into the adductor muscle.

The general appearance of the yellow pustules is shown in Fig. 38A. Smears show

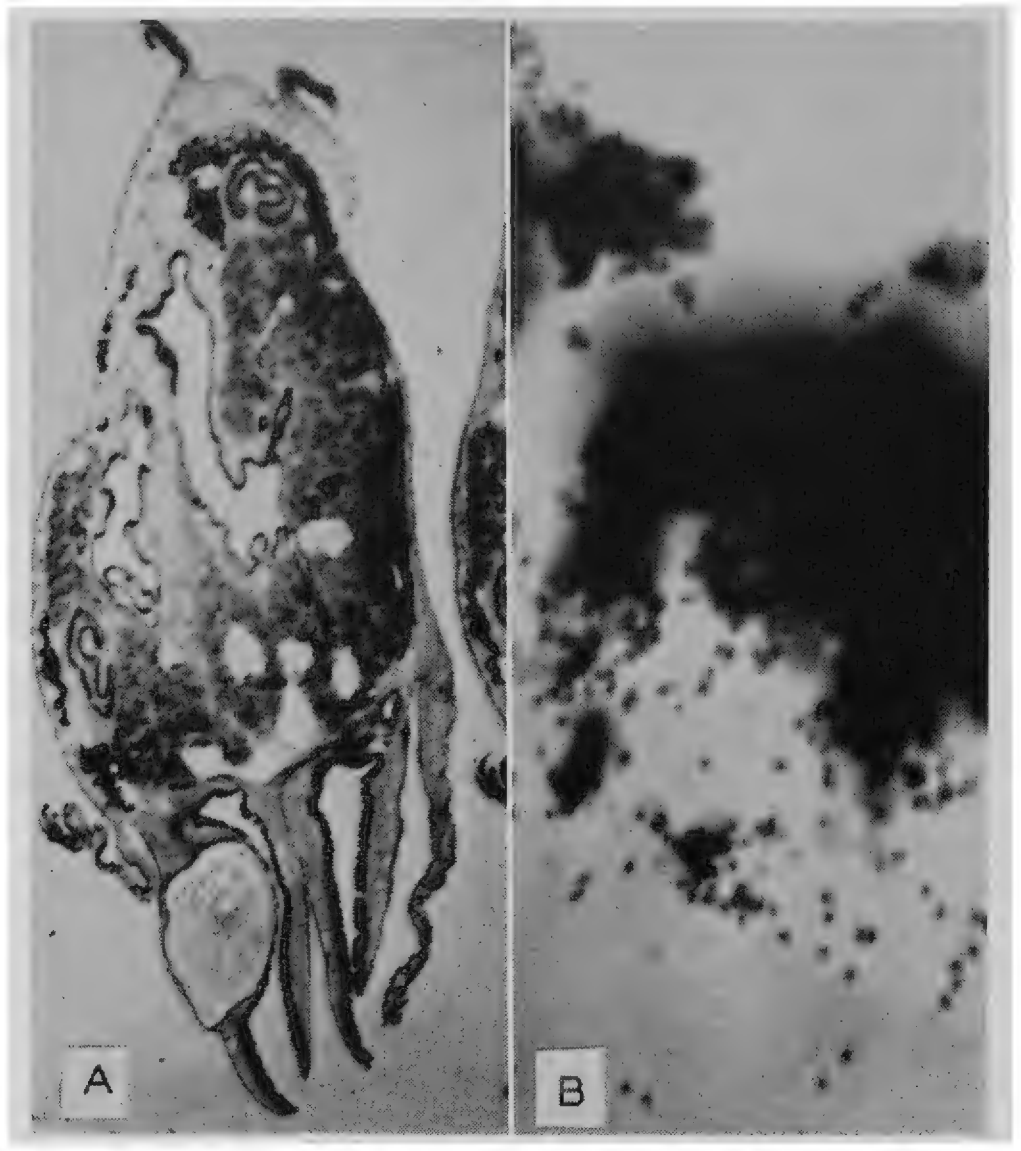

FIG. 37A. Section of oyster showing a watery cyst in the palp. This cyst is lined with epithelium. 37B. Bacterial smear from a watery cyst. The bacteria appear to be very small, short rods.

that they are filled with enormous numbers of moribund leucocytes in the cytoplasm of which are large numbers of basophilic granules. Stained with Giemsa these granules are purplish-blue and about 0.3 to 0.4 microns in diameter (Fig. 38B). They will also stain with iron haematoxylin and acid haematoxylin, and seem to be degeneration products. Bacteria may or may not be found in smears and may or may not develop on plates inoculated from abscess. Some abscesses seem to be sterile. In many cases leucocytes from absecesses contained inclusions of large size. These also do not appear to be parasites or to contain nuclei.

Bacteria when found in the yellow pustules are of varied type. None appear to be constantly present, and it is believed that most, if not all, are introduced saprophytes. The oyster appears to be capable of isolating and destroying these bacteria in most cases. Observations indicate that only a small percentage of all abscesses result in death of the oyster; these are usually associated with the adductor muscle, having been formed around mud introduced through *Polydora* burrows. The common "muscle pearls," nacreous excrescences in the muscle scar, represent healed abscesses in the muscle tissue.

The abscesses described here should not be confused with those caused by *D. marinum*. The latter do not have the yellow color. This yellow color is intensified in the bacterial pustules under ultraviolet light with a black light filter, which often shows the entire oyster to be a yellowish color, or makes certain large mantle arteries show as yellow lines. The normal color of oysters viewed by ultraviolet light is a sky blue.

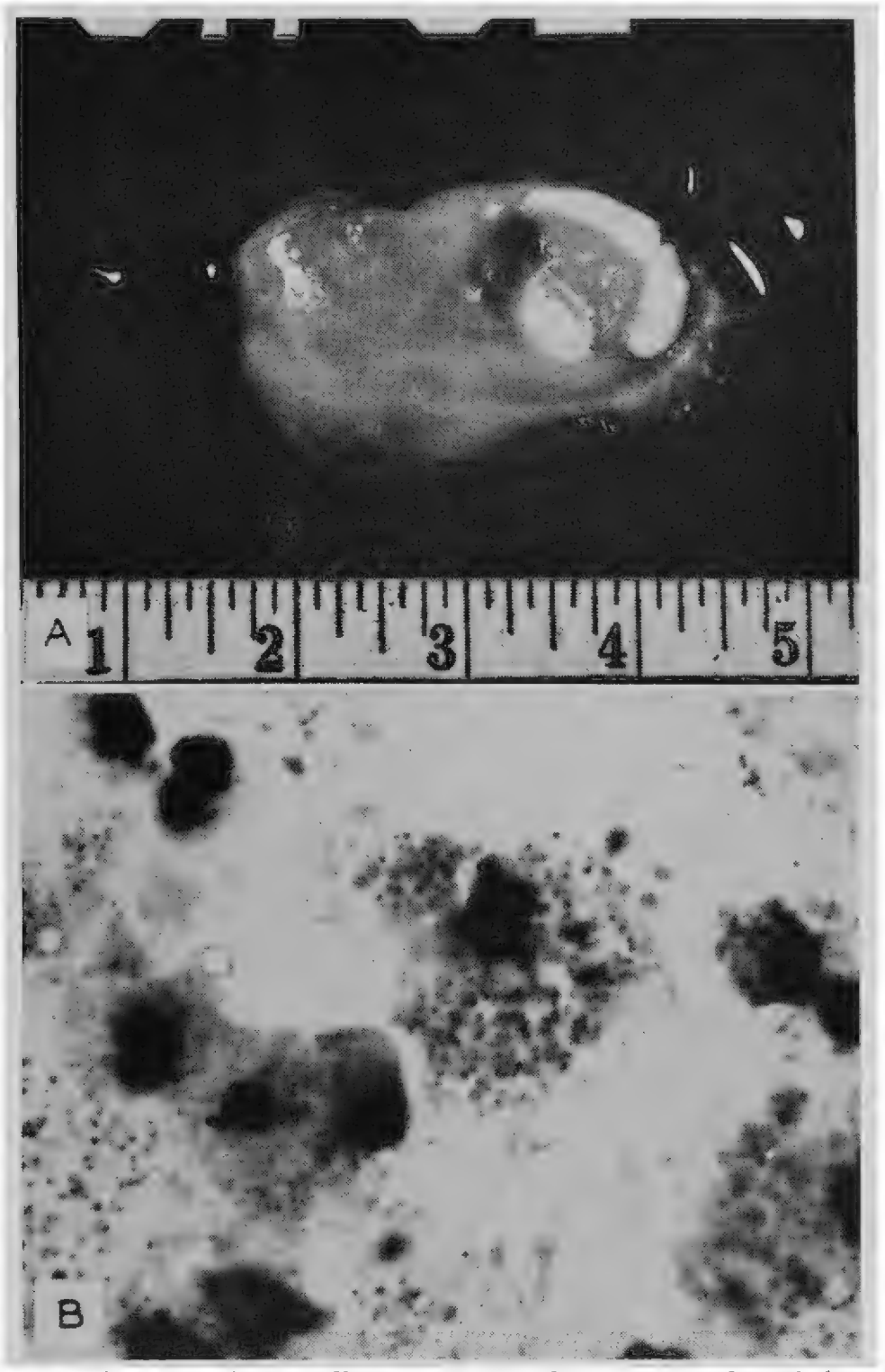

FIG. 38A. Oyster with a very large yellow pustule in the anterior edge of the adductor muscle. Note the thickened appearance of the mantle vessels under conditions of disease. 38B. Amoebocytes of the oyster from a yellow pustule. The small spherical inclusions are characteristic; stain with basic stains.

Yellow pustules do not ordinarily pose a great economic problem. Few oysters are marketed when the pustules are most common, in the high temperature period of late summer. Most oysters are thin and low in glycogen in that period, and by the time they are fattened in late fall most of the pustules have disappeared. Isolation by fibroid capsules and by epithelium formed from massed leucocytes often takes place, followed by sloughing of the entire pustule. These cast-off pustules may occasionally be found in the shell cavity, and may be covered with thin layers of shell by the mantle.

CILIATE PARASITE OF OYSTERS

A species of ciliate parasite of oysters is distributed over the Atlantic and Gulf coasts of the United States. Richardson (1938) mentioned a ciliate protozoan from Charlottetown, Prince Edward Island, Canada, which parasitized oysters and which he placed

in the genus *Orchitophrya*. If this parasite is the same as the species here described, Richardson's placement would seem to be in error. Richardson was able to transmit ciliates from host to host.

The ciliates are about 12 to 15 microns wide and 40 to 50 microns long. The macronucleus is ovoid and massive. The micronucleus occupies a vesicle either anterior to or posterior to the macronucleus but is never lateral to it. The anterior end is pointed, and encased in a long sheath is a needle-like retractile stylus with which it fixes itself to the digestive epithelium of the host. The ciliary rows are slightly spiral, about 14 or 15 in number. Contractile vacuoles are ordinarily not observed, but sometimes there may be up to three very small ones distributed from near the anterior end to the posterior extremity. This parasite probably is the same as *Ancistrocoma pelseneeri*, a widely distributed parasite of species of *Mytilus*.

This parasite is usually rather rare in healthy oysters, but in diseased oysters it is often found in large numbers in the stomach or intestine, and may penetrate the ducts and tubules of the digestive gland. It may become a complicating factor in *Dermocystidium marinum* infections but there is no reason to think that it has much independent effect on the host.

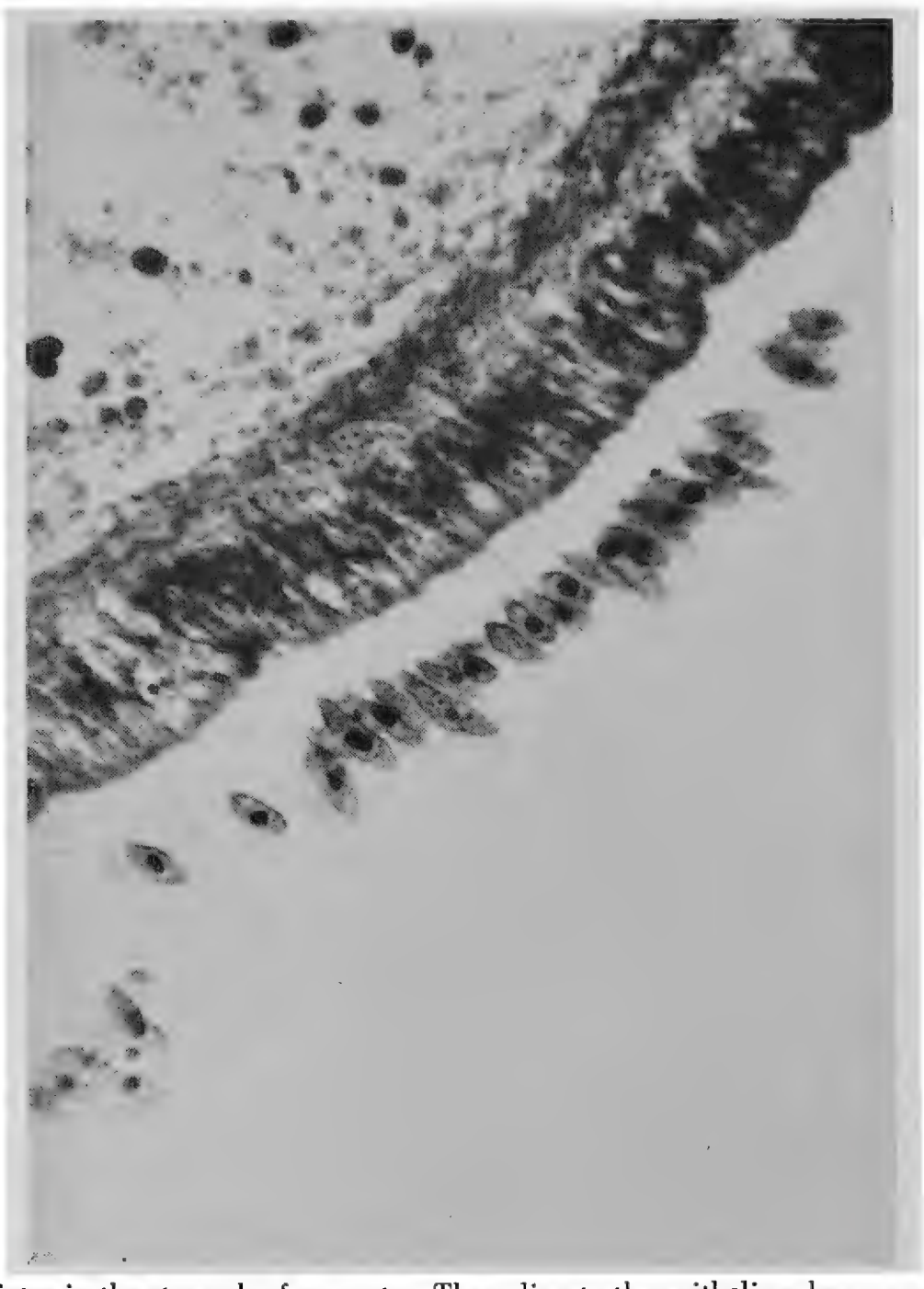

FIG. 39. Ciliates in the stomach of an oyster. They cling to the epithelium by means of a spear-like structure which can be retracted.

Fig. 39 is a photomicrograph of parasites in the stomach of an oyster. The row of ciliates here were originally attached to the epithelium but were detached by shrinkage due to fixation.

Acknowledgments

Studies of disease in oysters involved a considerable part of the personnel of Project Nine stationed at Grande Isle and all of the staff of Project Twenty-three. Full acknowledgment and an account of project administration is given in the preface to this volume. The author is especially grateful for assistance by Mr. Dan Wray, who accomplished much of the field work and made many slides for study in 1948 and 1949. Mr. James L. Boswell assisted in these studies from 1950 to 1960, and has accomplished much of the technical work during that period. Dr. Sewell H. Hopkins had advised in the work since the beginning of the studies.

Appendix

This appendix includes records of distribution of *D. marinum* in Louisiana collected by the Texas A & M Research Foundation Laboratory at Grand Isle, Louisiana, 1948 to 1959. Locations are listed in five regions; arranged from east to west. Bays and bayous are arranged generally in their order of location beginning near Mississippi Sound east of the Mississippi River and following down the east side of the river, around the end of the Mississippi River Delta, back along the west flank of the Delta to the northwest thence across the Barataria, Timbalier, and Terrebonne basins, to the Atchafalaya River, with one record west of this river.

In the listing below, the bay or bayou named is underlined, the date of collection of the sample follows the name of the bay, followed by (1) the number of oysters in the sample, (2) the incidence of infection in percent, (3) the weighted incidence of infection, (4) oyster lease number if known.

If a sample is too small for computation of incidence and weighted incidence, the location is given only as a positive identification of *D. marinum*, along with the date if known. After the underlined name of a bay, bayou or location, the samples in that location are continued to the next semicolon. If more than one sample is collected in the same bay or bayou on the same date, the data for the individual samples are separated by a "+" sign. Determinations based on gapers are indicated by the letter (G) at the end of the entry, all others are based on live oysters.

Superscripts refer to the following: a, See station 1 records (Mackin and Sparks, 1962); b, station 7 records, ibid; c, station 2 records, ibid; d, station 3 records, ibid; e, station 5 and 6 records, ibid; f, station 7 records, ibid; g, Mackin and Wray (1952).

I. *Area east of the Mississippi River; Mississippi Sound to Alexis Bay.*

Bayou Marron, 7–12–58/25[(1)]/0.0[(2)]/0.0[(3)]/14529[(4)]; *North Lake of Bayou Marron*, 7–12–58/25/0.0/0.0/14590; *Black Bay*, 8–24–51, at Stone Island, Gallego Island, and Lonesome Island; *Bay Craba*, 8–23–51, at S.E. end of Bay Gardene, 1–20–59/25/12/0.10; *Long Bay*, 8–23–51, east end, 3–10–50/6/17/0.77/8269; *Uhlan Bay*, 6–9–59/10/0.0/0.0 + 10/50/0.9 + 10/0.0/0.0, 9–3–59/2/0.0/0.0 + 10/0.0/0.0 + 5/40/0.3 + 5/

40/0.4 + 10/0.0/0.0; *Julius Bayou*, 2–5–59/40/40/0.38/13556; *California Bay*, 6–29–48, at California Point, 8–23–51, W. end of Bay, Middle of Bay; *Bay Denessee*, 9–10–58/25/40/0.64 + 25/44/1.16 + 25/36/0.68; *Old Stump Bay*, 7–3–59/30/53.3/0.90 + 7–3–59/2/50/2.5 (G); *Grand Bay*, 3–6–59/20/40/0.58, 12–7–58/10/50/0.75 + 10/30/0.20 + 15/73.3/1.77; *Drunken Bayou*, 12–14–57/25/48/0.60, 7–24–58/50/20/0.22; *Alexis Bay*, 7–24–58/30/26.6/0.57.

II. *In area around the end of the Mississippi River; active Delta area.*

Redfish Bay, 3–20–57/20/20/0.20 + 25/56/0.92, 7–3–53/45/0.0/0.0, 7–14–53/25/0.0/0.0 + 17/0.0/0.0, 7–15–53/16/0.0/0.0 + 12/0.0/0.0, 4–14–54/25/8.0/0.25; *West Bay*, 10–25–55/13/0.0/0.0 (G); Tiger Pass, 12–2–52/5/0.0/0.0.

III. *Spanish Pass to Grand Bayou; west flank of the Mississippi above the active Delta.*

Cane Bayou, 5–15–57/22/80/1.60; Spanish Pass, 5–15–57/20/55/0.95 + 25/24/0.16; *Bay Tambour*, 10–18–49/4/50/0.50, 8–12–53/25/32/0.36/11728, 8–10–57/19/0.0/0.0/14526, 8–28–58/25/0.0/0.0/14526 + 25/0.0/0.0/13628 + 25/4.0/0.12/14526; *Sandy Point Bay*, 8–11–53/25/88/2.20/11725 + 24/33/0.60/12988, 8–12–53/25/72/1.92/11213, 8–10–57/13/0.0/0.0, 8–28–58/25/8.0/0.06/13695; *Bay Jacque*, 10–18–49/4/100/3.0, 8–17–53/25/72/1.92/10455 or 10456, 8–18–53/25/60/1.34/9851; *Bay Chi Charas*, 8–26–53/25/84/1.18/12627; *Chi Charas Bayou*, 3–30–59/25/12/0.14; *Bay Coquette*, 6–25–51/1/–/1.0 (G), 8–23–53/25/36/0.50/11325 (G); *Cyprian Bay*, 7–3–51/1, Fasterling lease 262, 8–26–53/25/76/2.36/10762, 8–27–53/25/76/1.86/10164, 9–5–53/25/16/0.28/13100, 7–27–53/25/92/2.10/10464 + 25/76/1.34/12752, 7–28–53/24/75/1.17/12221, 7–29–53/21/67/1.17/9212; *Bayou Cop-Cop*, 8–31–53/25/76/1.10; *Dry Cypress Bayou*, 9–5–53/25/76/1.84/12751; *Pizatta Bay*, 1–31–55/25/36/0.26 + 20/35/0.25; *Bay Skipjack*, 10–18–49/6/50/2.16, 7–2–51/1, Vodopija lease 659, 8–27–53/25/92/2.50/10453, 8–28–53/25/84/2.28/10453; *Scofield Bay*, 7–19–51/1/–/5.0/625 (G); *Scofield Bayou*, 5–3–56/25/64/0.62, 8–29–51/1/–/5.0/635 (G); *Bay Pomme D'or*, 12–1–55/19/63/0.71/17600, 7–3–51/1, Fasterling lease 570, 8–19–53/24/67/1.23, 10–18–49/4/75/1.25; *English Bay*, 10–18–49/10/40/1.00, 7–3–51/1, Emmett Hingle lease 320, 8–27–53/25/88/2.56/11831; *English Bayou*, 9–3–59/34/35/0.62; *Fasterling Cut*, 9–5–53/25/76/1.60/11858; *Bayou Trouve*, 8–26–53/25/92/2.72/10446; *Bayou Fontanelle*, 8–29–51/1/–/5.00/10661 (G), 7–4–51/1/–/3.00 (G); *Bayou Borne*, 9–5–53/25/68/1.66/10741; *Bastian Bay*, 12–1–55/20/55/0.75/10046, 9–6–56/6/0.0/0.0, 10–18–49/8/63/1.62; *Bayou Cook*, 10–18–49/6/67/1.33; *Little Bay Cherie*, 11–1–55/–/–/0.82, Frank Jurisich, near upper Adams Bay; *Adams Bay*, 1–26–56/25/64/0.96/11863, 10–18–49/4/100/1.00; 11–30–55/4/0.0/0.0/11863 (G), 7–19–51/1/–/5.00/833 (G); *Bay de la Cheniere*, 12–14–54/15/13/0.33; *Bay Lanaux*, 9–24–58/40/22.5/0.45/13420, 2–7–52/26/50/0.54; *Grand Bayou*, 6–20–56/50/60/1.07, 5–28–57/23/65.2/1.13 + 22/31.8/0.80, 6–20–56/2/50/2.50 (G).

IV. *Grand Bayou to Bayou Lafourche.*

Lake Washington, 5–17–55/25/84/1.28 + 24/42/0.94; *Lake Robinson*, 12–16–59/33/12/0.12; *Bay Catherine & Bayou*, 11–27–56/25/44/0.72/13283; *Billet Bay*, 2–5–59/40/42.5/0.55, 8–7–58/60/55/0.65, 12–30–53/25/72/0.88/12861 + 25/84/1.36 + 25/80/0.94, 2–5–54/25/72/0.86/12861, 5–11–54/25/68/0.98 + 25/52/1.07, 5–25–54/25/80/1.27, [a], 1–24–57/20/75/1.12; *Freeport Sulphur Company dredged cut*, 1–23–57/25/52/1.08; 2–8–57/20/75/1.40, [b]; *Bayou La Mer*, 2–5–54/15/80/1.20; *Rattle-*

snake Bayou, [c]; *Grande Ecaille,* 10–8–49/21/52/0.76, 11–27–49, Eastern Grande Ecaille, 12–21–51, Western Grande Ecaille, [d], 1948–49/13/100/5.00 (G); *Four Bayou Bay,* 3–2–56/25/68/1.44/13140; *Bay Baptiste,* 7–20–49, Zibilich lease, 1949–50/27/100/4.70 (G); *Barataria Bay,* 10–16–54/25/68/0.92/13389 + 20/50/0.45/9457 + 11/55/0.95/9728 + 25/52/0.92, 10–18–54/25/60/0.78, 10–25–54/25/64/1.06, 3–21–55/25/40/0.50/9571, 3–28–55/25/44/0.62, 3–30–55/25/16/0.24/9942, 3–31–55/25/52/0.58/12887 + 25/44/0.85 + 25/52/0.62/12887, 4–22–55/25/56/0.92, 8–31–55/15/–/0.77/12899 + 10/–/0.25/12899 + 15/–/2.17 + 10/–/1.55 + 10/–/1.20, 8–19–49/31/35.5/0.55, 10–5–49/23/52/0.87, 4–3–50, 9578, W. of Queen Bess Island, near Bird Island, 8700. near Mendicant Island (S.W.), 8120, S. Barataria, 9557, Grand bank (S.E.), 1–23–51/8/12.5/0.125/8902, 2–8–51, 8902, Middleground, 3–21–51, West of Big Island, 9–15–53/25/40/1.02/10764, 3–28–57/13/61/0.88, [g], 8–31–55/6/–/3.1, May to Aug. 1949/25/100/5.00, 10–5–49/15/100/5.00, 11–8–49 to 4–22–50/15/93/4.66 (G), Sugar House Bend, [e]; *Sugar House Bayou,* 5–30–51/7/14/0.14, 1–15–57/25/80/1.26, [f]; *Bay Chene Fleur,* 1–20–54/1/0.0/0.0, 1–6–55/25/80/0.90, 1–5–51/25/40/–; *Little Bay Chene Fleur,* 2–24–60/15/0.0/0.0; *Wilkinson Bayou,* 10–6–49/26/0/0.0, 1–5–51/50/40/0.64, 2–7–51/20/15/0.15, 3–3–51/20/5/0.05, 4–4–51/20/20/0.30, 5–9–51/20/5/0.25, 6–5–51/20/15/0.15, 7–6–51/20/10/0.10, 8–7–51/20/20/0.35, 9–4–51/20/35/0.45, 3–22–52/44/24/0.29, 4–16–52/20/0/0.0, 6–25–52/25/0/0.0, 9–30–52/25/28/0.32 + 25/72/1.50, 1–14–53/50/50/0.72, 7–20–53/25/40/0.48, 4–20–54/25/76/1.92, 1–6–55/25/80/0.90, 1–9–56/25/40/0.52, 1–9–59/25/32/0.24, 3–3–59/50/6.0/0.15, 12–18–59/25/32/0.24, 2–4–58/25/24/0.16, 2–24–60/50/0.0/10.0; *Hackberry Bay* (Bay Du Chien, N.W. of Barataria), 8–4–52/15/20/2.00/10864 + 15/0.0/0.0/10186; *Manilla Bayou,* 5–1–56/47/4.0/0.04 + 37/0.0/0.0; *Bassa Bassa,* 10–7–49/21/38/1.33, 10–7–49/17/88/3.59 (G); *Creole Bay,* 10–26–49, 10364; *Bay des Islettes,* 1–7–54/25/84/0.60/13205; *Bayou Rigaud,* 10–3–49/21/67/1.14, Research Found. Bayou Rigaud Lab., May, 1949/3/33/0.33, Oct., 1949/21/67/1.14, April, 1950/6/33/0.33, June, 1950/7/86/2.07, Feb., 1951/20/40/0.60, Mar., 1951/20/10/0.10, April 1951/20/45/1.00, May, 1951/20/30/0.30, June, 1951/20/65/1.45, July, 1951/20/90/1.70, Aug., 1951/20/85/1.45, Sept., 1951/20/80/1.40, 4–4–52/25/52/0.88, 8–21–52/10/100/2.20, 3–4–52 to 5–31–52/279/70/1.35, 8–29–52/14/93/0.96, 9–26–52/25/56/0.64, 9–29–52/25/76/1.56, 3–4–53/25/52/0.72, 3–25–53/25/84/1.78, 4–13–53/25/88/1.90, 5–6–53/25/96/1.70, 10–10–58/25/76/1.36, 1–6–59/21/81/2.33, 2–6–59/8/25/0.38, 6–4–59/5/40/0.40; *Bayou Rigaud,* 1948–49/21/95/4.38 (G); *Caminada Bay,* 6–26–51, 9485, 10355, 10769, 8725, 9395; *Bayou Souris,* 1–10–55/25/36/0.26; *Bayou Thunder Von Tranc,* 12–28–53/5/100/0.90; *Bayou Fer Blanc,* 4–25–57/18/78/2.08 + 4/75/1.13 + 25/96/2.52; *Bayou Laurier,* 3–5–56/25/76/0.72/12823; *Bayou Lafourche,* 1–19–54/25/28/0.20.

V. *Bayou Lafourche—West.*

Panama Canal, 1–19–54/25/60.0/1.04; *Little Lake,* 7–12–58/50/52/0.70, 2–27–59/25/20/0.24, 3–25–59/26/12/0.08 + 26/54/0.71, 10–29–53/25/64/1.84; *Deep Bayou,* 10–29–53/25/80/1.72; *Bayou Blue,* 7–20–56/25/72/1.44 + 25/56/1.06, 8–10–51, (South of Little Lake); *Lake Raccourci,* 3–14–58/25/40/0.58/13883, 10–18–58/25/60/0.82/13883, 4–9–52/11/90.0/2.0 + 19/52.6/1.37; *Bay De Millieux,* 3–12–56/25/76/0.84/12429 & 12106, 11–1–55/25/48/0.58/10370; *Bayou Mulet,* 11–8–55/25/48/62/14191; *Bayou Rosedaux,* 11–2–55/25/72/0.92/12429, 11–9–55/25/76/1.30/12429,

11–10–55/25/52/1; 12/12429; *Bay Rosedaux*, 11–14–55/25/76/0.98/13333; *Bayou Demi John*, 3–12–56/25/68/1.08/11323; *Little Bay Rosedaux*, 3–12–56/25/56/0.64/10100; *Bay A Taton*, 2–15–56/25/56/1.02, 2–18–56/25/60/1.10; *Chinaman Bayou*, 11–3–53/25/56/1.80; *China Bayou*, 11–14–55/25/56/0.62/9400, 11–14–58/25/24/0.22; *Bay Courant* (Bay Chinois), 3–6–56/25/60/0.72/11916, 3–8–56/25/68/1.20/12219 & 12191; *Bayou Courant*, 3–8–56/25/88/1.14/11327 + 25/80/1.24; *Bay Plaissance*, 2–19–56/25/44/0.66; *Bayou Platte & Grosch*, 3–6–56/25/80/0.96/12822 + 20/60/0.60; *Bayou Grosch*, 3–6–56/25/68/1.12/13330; *Bayou Du Nord*, 3–2–56/25/76/1.08/12824 + 25/68/0.94/12824; *Bay Marselin*, 2–27–56/25/80/1.36/12422; *Bay Tambour* (above Raccourci), 2–28–56/25/52/0.58/12568 + 25/56/0.82; *Pass A John*, 6–21–58/70/65/1.56; *Bay Gray* (Grey), 2–28–56/25/72/1.08/12233, 7–4–58/15/87/1.60 + 10/70/1.75 + 2/50/0.26; *Deep Lake* (Lake Creaux), 2–27–56/25/76/1.16/10815 + 25/92/1.48, 11–3–53/25/52/1.28; *Middle Bayou*, 2–28–56/25/68/1.04/11748; *Bayou Du Chien*, 3–1–56/25/28/0.26/10518, 3–2–56/25/64/0.54; *Bayou Point Au Chien*, 1–14–58/25/24/0.20; *Timbalier Bay*, 10–25–55/25/84/1.56, 10–15–52/12/75/1.50, 1–19–54/25/68/1.26; *Grand Bayou Felicity*, 10–15–52/12/50/1.05; *Lake Felicity*, 11–3–53/25/76/2.64; *Lake Chien*, 11–4–58/30/40/0.67; *Bayou Dau Haute*, 4–23–59/10/30/0.45 + 5/40/0.30 + 5–20–0.60 + 5/20/0.10 + 5/20/0.80 + 5/60/0.40 + 5/60/1.00 + 5/60/0.40 + 5/20/0.20 + 5/40/0.30, 7–15–59/5/0.0 + 5/0.0 + 5/0.0 + 5/0.0 + 5/0.0 + 5/0.0 + 5/20/0.10 + 5/0.0 + 5/0.0 + 5/0.0 + 5/0.0 + 5/0.0; *Jacko Bay*, 10–29–53/25/76/2.06; *Lake Barre*, 8–10–51, (at Barre Oil Field); *Bayou Bas Bleu*, 10–21–49, (Research Found. planting); *Bayou Bourbeux*, 11–12–53/25/72/1.56; *Petite Caillou*, 10–6–54/25/100/1.88; *Bay Ste. Elaine*, 8–10–51, (at Ste. Elaine camp); *Rat Bayou*, 12–1–53/25/68/1.04; *Bay La Mat*, 12–1–53/25/68/0.76; *Bayou Billiot*, 12–1–53/25/60/0.86 + 25/36/0.24; *Moss Bay*, 1–12–54/25/56/0.76; *Oak Bayou*, 12–3–53/25/72/1.36; *Deer Bayou*, 12–3–53/25/56/1.12; *Lake Pelto*, 2–21–54/25/68/0.62 + 25/60/0.82; *Round Lake*, 2–21–54/25/60/0.64; *Dog Lake*, 12–4–53/25/92/2.12; *Bayou Du West*, 2–3–54/25/56/0.78; *Hackberry Bay* (West of Lafourche), 12–14–56/20/45/0.75, 12–8–49, South portion; *Bayou East of Hackberry Bay*, 12–4–53/25/72/1.44; *South end of Bayou connecting Dog Lake & Hackberry Bay*, 12–4–53/25/84/2.02; *Bayou between Hackberry Bay & Dog Lake*, 12–4–53/25/76/2.08; *Sister Lake*, 1–9–52; *Lake Mechant*, 12–16–52/40/17/0.17 + 35/14/0.14; *Mud Hole Bay*, 9–5–56/16/93.7/2.12/11315, 2–20–56/25/64/0.98/11315 + 25/52/1.08/9827; *Bannan Bay*, 2–3–54/25/40/0.66; *Oyster Bayou*, 2–18–60/40/7.5/0.05; *Tributary of Oyster Bayou*, 2–18–60/30/10/0.08; *Junop Bay*, 2–3–54/25/56/0.46; *Atchafalaya Bay*, 6–2–55/25/0.0/0.0, 7–12–53/15/0.0/0.0, 5–5–54/25/0.0/0.0 + 25/0.0/0.0, 7–15–54/25/25/0.0/0.0, 11–28–50/5/0.0/0.0 + 8/0.0/0.0; *Little Constance Bayou*, 11–25–53/25/72/0.66.

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Asterisks (*) indicate unpublished reports in the depositories of the 28 libraries listed in the preface to this volume.

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A Study of the Effect on Oysters of Crude Oil Loss From a Wild Well[1]

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Abstract

An oil well went out of control, caught fire, and spilled oil for two weeks in the Louisiana marshes in 1956. A series of stations was established with oyster trays and growth panels at varying distances. Measurements were made in the ensuing two years.

Mortalities of oysters in the area were primarily associated with incidence of infection by the fungus *Dermocystidium marinum* typical of Louisiana and were not related to distance from the well. Disease incidence, oyster set, growth, associated organisms on the reefs, and oyster conditions were usual for the Louisiana area and apparently not affected by the spillage. Oily taste in oyster meats could not be identified after two months.

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[1] Oceanography and Meterology Series from the Department of Oceanography and Meterology, and A. and M. College of Texas. A report from Project 23E, Texas A. and M. Research Foundation, sponsored by Texaco Incorporated, Humble Oil and Refining Company, The California Company, Gulf Refining Company, Phillips Petroleum Company, and Shell Oil Company.

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Introduction

The Oil Loss

On about November 17, 1956, an oil well, the Ernest Cockrell No. 40, drilled by Mecom Oil Company, went out of control, caught fire and spilled oil for a period of about two weeks. The exact period of the loss is not known to the authors. It is believed that the oil loss was probably the largest ever sustained in the oyster producing area of Louisiana. This accident provided an opportunity to set up a large scale experiment designed to determine whether or not such oil might affect survival, growth, or setting of oysters under natural conditions. In the weeks that followed, stations were set up at which oyster growth and survival were studied and compared with control stations located outside of the spill area .This paper is a report of the results of these experimental comparisons.

The well was located in the Freeport Sulphur canal a short distance to the north of the Grande Ecaille mine of the Freeport Sulphur Company (map, Fig. 1). The oil spread in all directions, some moving north in the canal and thence through Rattlesnake Bayou into Grande Ecaille and east to Lake Washington. Some oil spread to the southeast into Billet Bay and contiguous waters. On November 30, the fire was extinguished and the well blew oil and water for several hours before being brought under control.

Mr. Ed Schrader investigated the extent of the oil spread on December 6, 1956. On that date, a film of oil was found in the northeast half of Lake Grande Ecaille and in Rattlesnake Bayou. This was in the form of a heavy slick. Rattlesnake Bayou was covered by a film of oil for some distance east and oil along the shore was noted. Thickness of the film could not be measured but it was more than an iridescence. Freeport Sulphur canal had a heavy coating of oil and the entire shoreline was saturated, particularly along the west side, and to the south of the well location. In the Freeport Sulphur mine area and to the east into the dredged cut the oil was very heavy and in the greatest concentration recorded. In the north branch of Rattlesnake Bayou leading to Lake Washington, there was a heavy concentration of oil on the surface of the water. Oil was found in small amounts along the entire shore of Billet Bay in small coves. Traces of oil were found on the west shore of Lake Washington, but none was observed on the east side.

On December 11, a further check was made by Mr. Ed Schrader, chemist with the Grand Isle Laboratory of the Texas A & M Research Foundation. At this time a slick was observed as far west as Saturday Island in Barataria Bay, with scattered patches to the west and north of Saturday Island. Here the greatest concentration was between Big Island and Saturday Island, and was continuous eastward to the west entrance of Lake Grande Ecaille. In Grande Ecaille there were patches of tarry oil but no continuous slick. There were additional patches in Bay Ronquille, with pockets along the shore, and oil streaks on the vegetation and on shells along the east shore were observed. Heaviest concentration of oil was encountered near the southwest entrance to Grande Ecaille, near its junction with Ronquille Pass. Along the south and east shores of Grande Ecaille was another heavy concentration, and a large tarry patch was contained in the cove adjacent to the Freeport Sulphur mine on the south and east sides of Grande Ecaille.

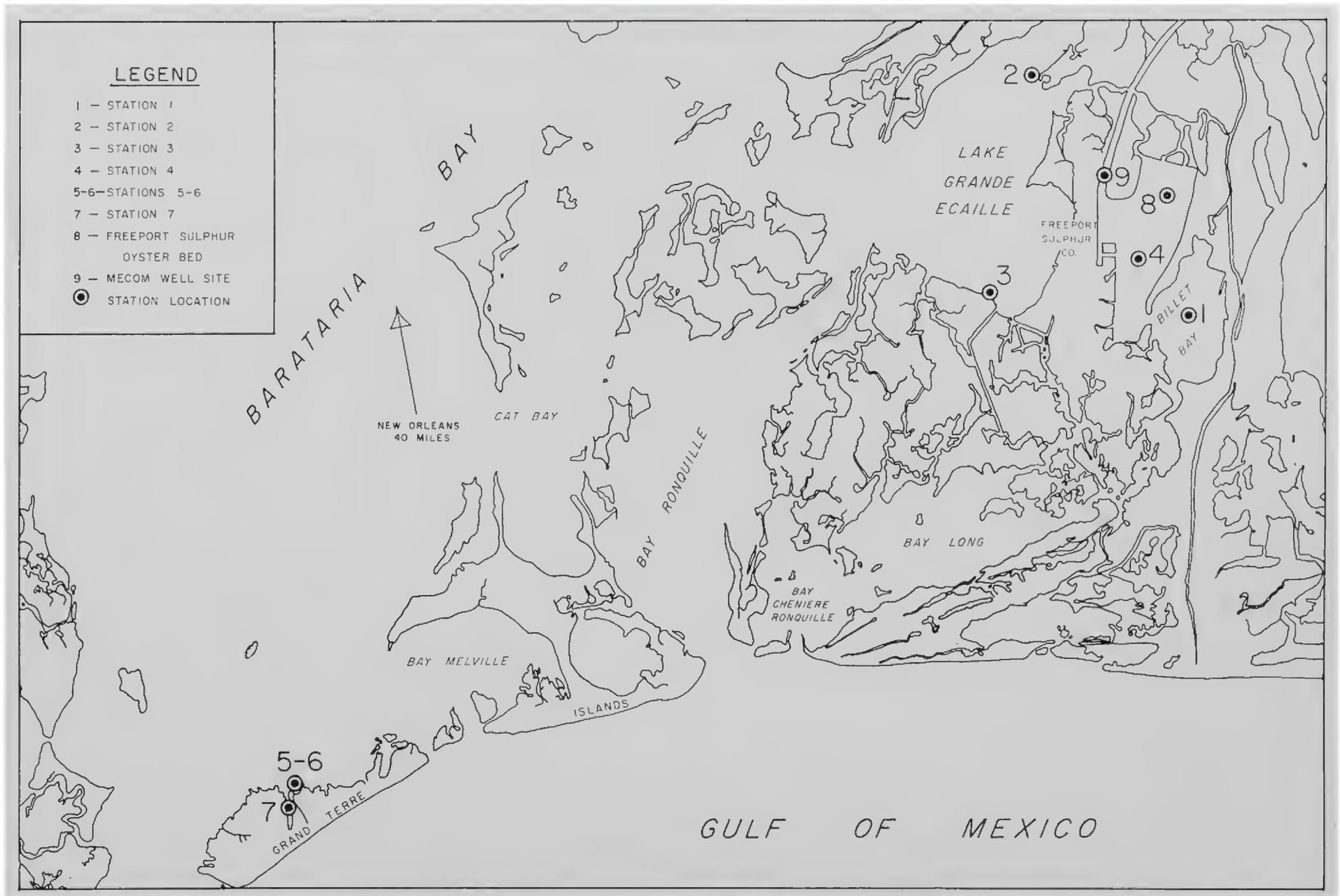

FIG. 1. Map of the area showing location of the wild well, the oyster bed in the dredged cut from which the oysters for this study were taken, and the locations of the seven field stations.

Oil in the Freeport Sulphur canal extended northeastward beyond the junction with Bayou Cheniere. Up to the junction with Bayou Cheniere there was much oil, but it thinned out to the northeast and disappeared about midway between Bayou Cheniere and the Grande Bayou crossing.

On December 13, the junior author observed that the oil in Barataria Bay had disappeared, and no oil was observed west of Grande Ecaille on that date except a light film near the west entrance. Rattlesnake Bayou was also clear, but there was considerable oil on the water and along the shorelines of the Freeport Sulphur canal. To the east, oil was observed in the pass between Billet Bay and Lake Washington, along the shorelines, and in marsh vegetation.

On December 26, 1956, the authors visited the area and, with Mr. Fred Deiler of the Freeport Sulphur Company, inspected the local area from Billet Bay into the dredged cut to the east of the Freeport Sulphur Company. No oil was observed in Billet Bay, but there was much oil in the dredged cut along the shores, but no more than light slicks over the surface. However, in tonging oysters on the Freeport Sulphur planted bed in the cut, oil was dislodged from the bottom and floated to the surface in considerable amounts. As late as February 5, 1957, oil could still be stirred from the bottom of this bed. Thus, the oysters used in stocking the experimental stations for this study were subjected to oil from the latter part of November, 1956, to February 5, 1957. It is not known just when oil from the burning well reached the bed after November 17, but the exposure to oil was certainly for more than two months.

Tonging oysters from the Freeport Sulphur Company bed in Billet Bay on December 26 failed to produce slicks. No bottom oil was observed outside of the immediate vicinity of the well and in the dredged cut, but it is assumed that around the margins of the bays some oil was absorbed on the mud.

In assessing the extent of the oil loss, there is no way to determine how much oil escaped. All light fractions must have been burned when the well was on fire and much more evaporated. Thus most of the lost oil was artificially "weathered" except in the short period of several hours after the fire was extinguished, but during which the oil flowed unhindered. In no case were the authors able to measure amounts of oil in any area with exactness, or to make exact estimates of thickness of oil film.

During the six months of the study, the west end of Rattlesnake Bayou was observed to have almost continuous oil slicks. These had no relation to the original oil loss but came from the intense area of oil development in the end of the bayou. While this oil apparently was not connected with the Mecom well, it has a part in the study, simply because it was there, and close to our experimental Station 2. Station 2, located just to the north of the junction of Rattlesnake Bayou with Lake Grande Ecaille, was almost continuously subjected to light slicks for the duration of the study. It thus became a master experimental station.

Station 2 also inadvertently became an experimental station in another sense. In the latter part of February, a canal was cut by Humble Oil and Refining Company just to the west of the racks. Oysters had already been placed at this station, although the formal checks had not begun. No effect from turbidity produced by this dredging could be observed. No silting of the oysters occurred.

Disappearance of the Oil

Oil disappeared very rapidly from the surface of the water in the bays and bayous, but occasional slicks which appeared to come from pocketings in the marsh and along-

shore were seen to the end of December, 1956. Thereafter discolorations on the marsh plants and on shells along the shore gradually faded. By the time the actual data-taking began in early March, 1957, physical appearances were normal except in the immediate vicinity of the well and in the dredged cut of the Freeport Sulphur Company.

The floating oil was flushed to the Gulf, the major part probably passing through Bay Ronquille and Bay Long and out through Quatre Bayou Pass into the Gulf. A minor part probably was discharged through Chaland Pass, and it is possible that some oil slicks went through Pass Abel. However, no slicks were ever observed in Lower Barataria Bay, although frequent trips were made along the southern and eastern shores. It is believed that the oil observed in northeastern Barataria Bay was discharged through Cat Bay and thence through Quatre Bayou Pass.

Much oil was evaporated as evidenced by the weathered appearance of the floating patches which were observed.

Remaining oil was reduced by bacterial oxidation. Prokop (1950) has studied bacterial decomposition of petroleum and has shown that oil cannot be retained by muds for long periods of time. Brown (1950) has also studied the bacterial reduction of crude petroleum and Brown, Van Horn, and Reid (1950) demonstrated that microorganisms in Louisiana muds destroy a large number of the compounds of crude petroleum. ZoBell (1949b) also measured the rate at which oil is oxidized by bacteria, and he furthermore (1949a) showed that bacteria capable of destroying oil exist in Louisiana bay muds. The oil oxidizing bacteria were found only in small numbers, but increase rapidly to large numbers under conditions of contamination with crude oil. They were shown to be widely distributed, in bottom muds of Louisiana bays.

Data Collected, Methods, and Stations

The data taken in this study include the following:

(1) Measurements of the total mortality of oysters were made at several stations for the period of the study, and measurement of the rates of mortality were made during successive periods.

(2) Analyses of the effects of the fungus disease *Dermocystidium marinum* in production of mortality of the oysters were based on diagnosis by the culture method devised by Ray (1952a, 1952b). A total of more than 1000 gapers and 1878 live oysters was cultured during the study.

(3) Study of several other minor agencies producing mortality was also made.

(4) "Condition" in control and experimental oysters was measured, using the coefficient-of-fitness method.

(5) Studies were made of the macroscopic fauna of the trays, comparing control and experimental stations and the faunas of all stations with normal north-Gulf faunas as described by Hedgpeth (1953) and Gunter (1955).

The Stations

Seven stations were set up; Stations 1 through 4 were designated experimental stations because they were in the area of heavy oil contamination. Stations 5 through 7 were placed at a distance from the contaminated area in lower Barataria Bay and were designated control stations. Stations 5 and 7 were stocked with oysters which had been

subjected to oil, then removed to the uncontaminated control area. Station 6 was stocked with oysters which had never been in contact with oil. All stations were constructed in the last two weeks in February, 1957. Oysters were placed at the stations in the period of the last week in February and the first week in March. The last culling mortalities were removed on March 12 and the study began on March 13, 1957.

The stations of the experimental group were in a region where salinity is "high," varying generally from about 10 ppt to about 27 ppt.

At the three control stations in lower Barataria Bay the salinity range was from about one ppt to 35 ppt, a greater range but about the same median as that at the experimental stations.

Station 1 was located in Billet Bay (Fig. 1) a considerable distance from the shore in any direction. Water depth at Station 1 was about three to four feet and the bottom was fairly stiff mud. The station was in an area of intense oyster production of probably an above average type for Louisiana.

Station 2 was in Lake Grande Ecaille just to the north of the junction of Rattlesnake Bayou. It was in an area of intense oil activity. Fig. 2 shows the rack and in the near

FIG. 2. Photograph showing the location of Station 2, an experimental station, in the northeast part of Grande Ecaille at the mouth of Rattlesnake Bayou. A christmas tree and drilling well of the Humble Oil and Refining Company are shown in the near background.

foreground to the left is a completed well and to the right a drilling well of the Humble Oil and Refining Company. Drilling at this location was in progress during most of the period of the study.

Station 3 was located on the south shore of Lake Grande Ecaille in about three feet of water. Heavy oil from the Mecom well was pocketed along the south and east shores of Grande Ecaille.

Station 4 was located in the dredged area to the east of the Freeport Sulphur Company mine. It was fairly close to shore, but in comparatively deep water. All other stations

were in three to four feet of water, but here the depth was about eight feet. The construction of this station differed from that of the others. The entire structure was iron or steel and the "baskets" were of heavy iron meshwork supported with an iron frame. These trays were hauled to the top of the platform by means of a hand-operated chain hoist. When submerged, each tray was covered with a rectangular iron meshwork on a frame.

Stations 5 and 6 were placed within 20 to 25 feet of each other. One of these (Station 6) was the "master control" and was placed as near as practical to Station 5 (Fig. 3).

FIG. 3. Photograph of the racks of Stations 5 and 6, two of the control stations. The marsh beyond is Grande Terre east of the entrance to Sugar House Bayou. The area is referred to as Sugar House Bend.

They were in the area called "Sugar House Bend," near the north shore of Grand Terre Island, in the southern part of Barataria Bay. The bottom at Stations 5 and 6 was packed mud-sand mixture, and the depth was three to four feet. This location was chosen because there are available extensive data on mortalities from studies carried out in previous years.

Station 7 was placed in Sugar House Bayou, which transects Grand Terre in a general northwest-southeast direction. Originally this "bayou" was a part of the extensive drainage system when attempts were made to grow sugar cane on Grand Terre. Station 7 differs from other stations in that it is in an area where natural populations of oysters grow in small reefs along the edge of the bayou. Natural populations of oysters do not grow in the area around the other stations, where the capacity for maintenance has long since been lost. Station 7 differs in one other respect from the other locations. In summer, water temperature is often very high during ebb tides, sometimes reaching 40°C (Mackin and Cauthron, 1953).

CONSTRUCTION OF THE "RACKS"

Wooden structures, made of creosoted "two by fours," were used to support the trays of oysters. They were joined together with galvanized bolts. The lower ends of the

uprights were sharply pointed and when the racks were installed the pointed ends were forced into the mud bottom until the cross stringers on which the trays rested were on the mud. Cat walk frames were built alongside the supporting frames for the trays, and joined with cross braces on which the cat walk planks were placed. These units are very strong; those used in this study passed through hurricanes Bertha and Audrey without damage and without losing an oyster.

The use of tarred trays for raising oysters is standard procedure in different parts of the world. The trays are usually supported on creosoted wood structures. For a description of a highly successful operation using the identical trays used in this study, see Anonymous (1940). Also see Nelson (1934) and Hewatt and Andrews (1954).

Trays were prevented from washing off the stringers by heavy wire bridles which were attached to the upper stringers. When the trays were examined they were lifted to the surface with the bridles. All trays were sealed by a second inverted tray which was hinged to the lower tray and wired in place before leaving at each visit. One of the trays is shown in Fig. 4.

FIG. 4. Trays containing oysters at Sugar House Bend. Note the galvanized rat wire lining used to exclude predators, and the board partition which was riddled by shipworms. Mr. Thatcher (left of the observer) and Mr. Landry check for gapers. Note the cat walks on which they stand. These boards were removed after each visit to discourage tampering.

OYSTERS

Oysters used for the four experimental stations and two of the control stations were taken from the Freeport Sulphur Bed in the dredged cut just to the east of the Grande Ecaille mine. The location of this bed is shown on the map, Fig. 1. The oysters ranged from seed size to quite large oysters (see size distribution diagram, Fig. 5). At the time they were tonged and dredged for distribution to the stations, they were growing well, were fat, and well shaped. As stated, this area was heavily contaminated with oil, and the mud still contained sufficient oil to form slicks when the oysters were taken up. They had been subjected to this condition for more than two months when removed to the stations.

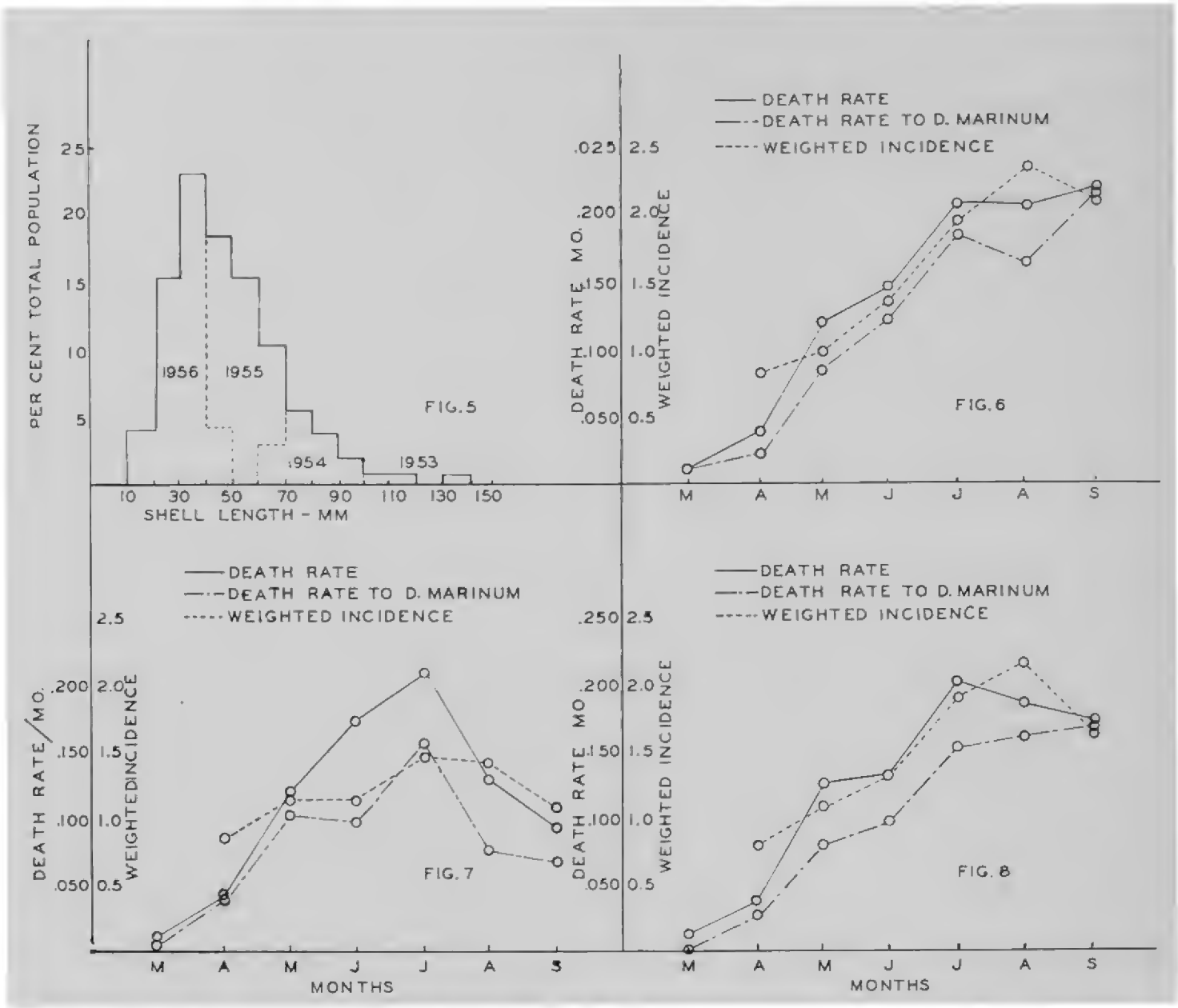

FIG. 5. Size distribution diagram for oysters in a sample from the Freeport Sulphur Company planting in the dredged cut. This histogram shows the sizes of oysters used in stocking Stations 1, 2, 3, 4, 5 and 7, from which the year classes and maximum age of the oysters present were determined.

FIG. 6. Death rates from all causes, weighted incidence of *Dermocystidium marinum*, and death rates from this fungus disease at Station 1, March to September, 1957.

FIG. 7. Death rates from all causes, weighted incidence of *Dermocystidium marinum*, and death rates from this fungus disease at Station 2, March to September, 1957.

FIG. 8. Death rates from all causes, weighted incidence of *Dermocystidium marinum*, and death rates from this fungus disease at Station 3, March to September, 1957.

Oysters used for the master control station were obtained from an oysterman in lower Barataria Bay. These had recently been transplanted from a low salinity area above Timbalier Bay just prior to the time they were obtained, a fact not known at the time they were purchased. They had a very low incidence of *Dermocystidium marinum* infection compared with the experimental oysters. This accident presented an opportunity to incorporate into the study a test of the theory that subjection to oil may enhance the effect of *D. marinum*, and that incidence of infection by the fungus is increased by the presence of petroleum.

The possibility that subjection to oil is necessary for production of *D. marinum* mortalities or development of epidemic disease has been tested with negative results (Mackin, 1950). However, no field tests have hitherto been carried out. We were, therefore, presented with an ideal situation, namely that of being able to compare develop-

ment of epidemic disease in oysters which had neither been subjected to oil nor to fungus disease, with oysters which had had the maximum field dosage of crude petroleum, came from an endemic area, and were infected at the beginning of the study.

Each of the seven stations was provided with trays which held 1000 oysters. At each station there were 300 oysters used for determination of total mortalities. This group was used for nothing else, except that gapers recovered were subjected to the Ray culture technique for determination of incidence and intensity of infection with *D. marinum*. Since only a small part of the meat was used in culturing, the remainder of the oyster was fixed in Zenker's fluid, when not too badly disintegrated, in order to allow for studies of other diseases should such appear.

Six hundred oysters at each station were reserved for analysis of intensity of disease caused by *D. marinum* in the live oysters. Samples of 50 oysters, 25 of them relatively large and 25 small, were withdrawn from this group each month for the measurement. All gapers from the group were also cultured and fixed to provide further checks on fungus disease in dying oysters. The group also contributed to the determination of mortality level from month to month.

The third group of oysters, numbering 100, was reserved for condition studies, and 10 were withdrawn for this purpose each month. Toward the end of the study the group dropped so low in number that the live oysters used for diagnosis of *D. marinum* were used for condition studies after excising the small piece of tissue used for the culture. No checks for condition were made at the beginning of the study. Condition was measured by the "index of condition" method (dry weight of meats in grams per shell capacity in cc, times 100). The volume of the shell cavity was determined by the displacement method and the meats were dried at 100°C to constant weight in a drying oven. A chainomatic balance was employed in the weighing.

Presentation of the Data

Mortality

Mortality rates for the region as shown by age analysis. Prior to the beginning of these studies a sample was taken from the bed of the Freeport Sulphur Company in the dredged cut (Fig. 1). This sample was taken on January 23, 1957, and is representative of the oysters used in these studies (excepting those at Station 6). The length of the right valve of the shell of each oyster of the sample was measured, and the size frequency plotted to provide data with which to calculate growth during the period of the study. An estimate of the annual mortality to which these oysters are subject may be made by examination of the size distribution diagram. The method consists of identification of the maximum number of year classes in the population, using the modes in the size distribution diagram and basic knowledge of growth rates gained from previous studies. The current year class may be separated from all older year classes on the basis of color and general appearance of the shell. This was done at the time of measurement of the oysters.

The size distribution diagram is shown in Fig. 5. The oldest oysters in the population probably set in the spring and summer of 1953, certainly not earlier than late April or May of that year. On January 23, 1957, they were, therefore, about 3½ years old. Only a remnant of that year class remained alive, since it made up only about two

percent of the oysters represented in the diagram. An average annual mortality of at least 60 percent is indicated, the figure necessary to reduce a population to such a low level in 3½ years time.

The steps in application of the method were as follows. First, it was necessary to know the size of oysters which are in their third year. This information was obtained from actual measurement of growth of oysters in previous studies and the sizes which they may attain. Reference is made to two representative studies. The first study was reported by Menzel (1950). He found that oysters at Bayou Bas Bleu grew from setting (in May) to a mean shell length of 58 mm (about 2⅓ inches) in one year. Oysters one year old grew in their second year from about 54 mm to about 84 mm (about 3½ inches). Oysters in their third year grew from 86 to 101 mm, at which time they were almost exactly 4 inches long (100 mm). All figures represent means.

At the end of three and one-half years the oysters in our sample, at the rate found by Menzel, should have grown to a mean size of 112 to 125 mm. In the second study Dawson (1955) got results similar to those of Menzel but did not check growth in the first six months. Ingle and Dawson (1952) found a higher rate of growth in Apalachicola Bay, Florida, but pointed out that their data indicated that in this bay growth of oysters was superior to any other area studied in the United States.

An examination of the size distribution diagram (Fig. 5) shows that only a few oysters of the sample from the dredged cut bed of the Freeport Sulphur Company exceeded 100 mm in length and none attained 140 mm in length. Thus from the sizes there were few, if any, oysters older than 3½ years in the sample. This conclusion is supported by study of the diagram itself. Each year class, if it could be separated from the whole would make a more or less symmetrical diagram (the "normal" curve). The 1956 year class was separated from the older oysters when measuring on the basis of shell color and form (Fig. 5), and there can be no question about the age of that group, which contained oysters four to eight months old. The oysters in the block to the right of the 1956 group must represent the 1955 class, which was up to 20 months old. Thereafter no definite division exists because of reduction of numbers by deaths and fusion of the modes by reason of slower growth in the second and third years. However, the marked asymmetry of the right hand limb of the histogram indicates that at least one other year-class (1954) is represented (2 to 2½ year old), and the few straggling oysters over four inches in length indicate that these probably were between three and four years old. Certainly the oldest year class represented is less than 5 percent of the number of the youngest year class: it is reasonable, therefore, to assume that this group had sustained more than 95 percent mortality since setting in 1953.

Table 1 indicates the period required to reduce a population to less than 5 percent of its original size with various mean percentages of annual mortality. At 10 percent mortality, more than 28 years would be required to reduce a population to less than 5 percent of its original size. With a known longevity of three and one-half years the mean annual mortality rate would be about 60 percent.

At the beginning of the study it was, therefore, indicated that if the time of the investigation carried over a considerable part of the summer, the mortality should average above 50 percent. Past studies in Lake Grande Ecaille (Mackin and Wray, 1950) showed that in the period from March to September the mortality was 63 percent and at Sugar House Bend, it was 74 percent in the same period.

TABLE 1

Approximate time required to reduce a year-class population of oysters to less than five percent of its original size at different mean annual rates of mortality

Mean annual mortality rate (percent)	Approximate number of years to reduce to less than five percent
10	28 to 29
20	about 13
30	8 to 9
40	about 6
50	4 to 5
60	3 to 4
70	2 to 3
80	nearly 2
90	1 plus

Mortality from the date of oil loss to the sampling date. The mortality rate derived from this method applied to oysters in the area from which the seed for planting the bed were taken, since too little time had elapsed after planting for the environment of the dredged cut to affect mortality rate. Indications were that very little mortality had occurred in the last two or three months before the beginning of the study. Recognizing that numbers of boxes (hinged valves) do not indicate mortality rate (Menzel and Hopkins, 1952), the condition of the boxes will show whether or not a recent rapid mortality has occurred, if that mortality was significant and concentrated in a relatively short period of time. The sample taken on January 23 had less than five percent boxes, none of which were "fresh", *i.e.*, not fouled. Even if all of the boxes had been "new" the mortality rate since the oil loss would have been negligible.

Cumulative mortality of oysters at the control and experimental stations, March 3, to September 4, 1957. Table 2 shows the total mortality for the seven stations used in this

TABLE 2

Mortality of oysters in percent at the experimental and control stations for the entire period of the study, March 3 to September 4, 1957. Data are from the oysters in the "mortality" trays only, 300 at each station, and from the total population beginning with 1000 oysters and calculated from the mortality rates in different periods as shown in Table 4

Station		Total mortality (using 1000 oysters) percent	Means	Total mortality (300 oysters) percent	Means
Area	No.				
In oil area	1	65.1		60.7	
	2	59.0		52.7	
	3	61.2		65.0	
	4	30.4		28.3	
			53.9		51.7
Control area	5	48.0		42.3	
	6	80.7		79.7	
	7	63.5		55.0	
			64.1		59.0

study. With the exception of Stations 4 and 6, the mortality rates were about as expected. Station 4 had distinctly less mortality than expected. However, the average (51.7 percent) for the experimental stations and the average (59.0 percent) for the controls were sufficiently in agreement that it appears that the differences were not significant. An overall difference of seven percent is slight considering that between experimental

stations there was a difference in total mortality of about 37 percent (Stations 3 and 4), a difference which was matched by that between two control stations (5 and 6) of 37 percent.

Mortality rates in the different periods (months) of the study. Total mortality figures do not show changing rates of mortality. All stations began with a relatively low rate of mortality which generally held through March and April (Figs. 6 to 12). Thereafter,

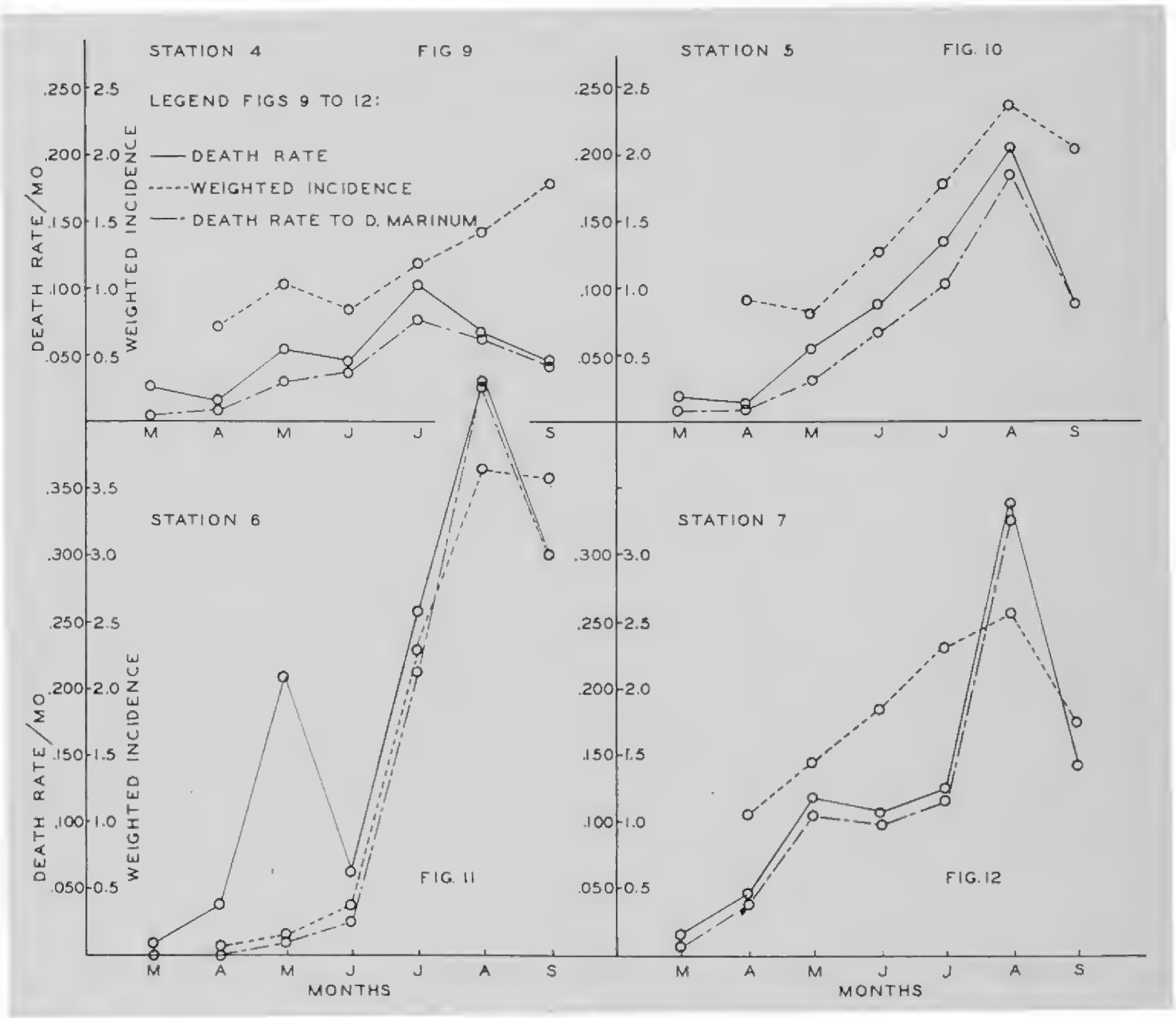

FIG. 9. Death rates from all causes, weighted incidence of *Dermocystidium marinum*, and death rates from this fungus disease at Station 4, March to September, 1957.

FIG. 10. Death rates from all causes, weighted incidence of *Dermocystidium marinum*, and death rates from this fungus disease at Station 5, March to September, 1957.

FIG. 11. Death rates from all causes, weighted incidence of *Dermocystidium marinum*, and death rates from this fungus disease at Station 6, March to September, 1957.

FIG. 12. Death rates from all causes, weighted incidence of *Dermocystidium marinum*, and death rates from this fungus disease at Station 7, March to September, 1957.

rates followed the temperature rise in the spring rather closely. The maximum rates were reached in either July, August, or September, depending on the station. At Station 6, the rate reached the very high figure of 481 oysters per thousand population in August, the highest rate ever observed in ten years of study in Louisiana. This was the master control station. It is believed that the tables and graphs give sufficient information so that further discussion is not necessary.

Causes of Mortality

As indicated above, all stations were established and attended in such manner that the causes of mortality could be determined with considerable accuracy. In Louisiana the causative agencies of most importance include (1) fungus disease caused by *Dermocystidium marinum*, (2) conchs *(Thais floridana)*, (3) crabs, especially the stone crab *(Menippe mercenaria)*, (4) certain fishes, the drum *(Pogonias)* and sheepshead *(Archosargus)*, (5) *Stylochus*, the "wafter" or "leech", a polyclad predator, (6) *Polydora websteri*, mud worm, (7) *Odostomia*, a gastropod parasite, (8) possibly boring sponge *(Cliona spp.)* and boring clam *(Martesia smithi)*, (9) diseases other than that caused by *D. marinum*, including disease caused by *Bucephalus*, and a "new" disease to be discussed below. Several other diseases of obscure nature are known to occur in oysters in Louisiana, but their effect cannot now be measured. They may, in the aggregate, be of importance. The stations were set up to exclude most predators, such as the large fishes and, to some extent, conchs. Small predators could not be excluded and all parasites could gain easy access to the oysters.

In addition to the parasites and predators, certain other factors of physical nature may kill oysters. Except for one case of mudding over, none of these were operative in the trays during this study.

Those agencies which were determined to be of importance in mortality are discussed next.

Disease caused by Dermocystidium marinum. Of all causes of mortality this fungus disease so overshadowed all others combined that most of our efforts were directed to measurement of its effects. This was expected since all previous studies in the area have shown its paramount importance.

The pathogenicity of *D. marinum* infections has been established by a number of studies. Mackin (1951) studied the effect of heavy infections on the tissues and described the destruction of the Leidig cell system, intestinal epithelium, muscle, and other tissues. Ray, Mackin, and Boswell (1953) measured reduction of weight of oysters parasitized by *D. marinum*. A series of studies involving experimental infection of oysters by *D. marinum* (Mackin, Ray, and Boswell, 1953; Ray and Mackin, 1954; Ray, 1954a; Ray and Chandler, 1955; and Andrews and Hewatt, 1957) showed that the disease is highly lethal when infections are in advanced stages (classed here as moderate, moderate to heavy, and heavy), while lighter infections apparently do not kill oysters.

The intensity of infections in individual oysters can be readily determined by the method developed by Ray (1952a, 1952b), and, as already indicated, this method was used in diagnosis and classification of intensity of infection in the present study.

Experience has shown that *D. marinum* infections cause a high percentage of the deaths which occur in tray oysters (Mackin, 1953; Andrews and Hewatt, 1957) in endemic areas. The latter authors found that, in oysters from endemic areas, 80 to 90 percent of the oysters dying had heavy infections of the fungus, although the winter percentage was much lower than that of the summer period. In Virginia, annual losses observed by these authors varied from 26 percent to 57 percent, of which 87 percent was attributed to fungus disease. Ray (1954a) found that uninfected oysters introduced into high salinity Louisiana waters contracted the disease to the extent of 90 to 100 percent in one month, of which about 80 percent died in a few months, with about 98 percent of those dying having heavy infections of the disease. Owen (1955) found that

the number of gapers with heavy infections of *D. marinum* was about 47 to 81 percent. His gapers were taken from 10 stations in Louisiana at which mortality rates of 26 to 80 percent in 8 months occurred. Similar results have been obtained by the senior author in several studies.

Analysis of effect of fungus disease in the present studies. Pre-study analysis. In January, February, and March several samples of oysters were cultured to determine the intensity of infection in the region of the oil loss and at the site of the control stations. One sample was from the Freeport Sulphur bed from which oysters were taken to supply all stations excepting Station 6. A summary of the results of these analyses is given in Table 3. The first three beds listed in this table were more or less subjected to oil con-

TABLE 3

A summary of results of analysis for incidence and weighted incidence of fungus disease of several samples of oysters in the Barataria area and Sugar House Bayou

Origin of the samples	Date, 1957	Number of oysters analyzed	Incidence of *D. marinum* (percent)	Weighted incidence
Freeport Sulphur bed in dredged cut*	1–23	25	52	1.08
Freeport Sulphur bed in Billet Bay	1–24	20	75	1.12
K. Tesvich bed in dredged cut of Freeport Sulphur Co.	2–8	20	80	1.40
Sugar House Bayou	1–15	25	80	1.26
K. Tesvich bed in Barataria Bay	3–28	13	61	0.88

* All oysters for supplying all stations except Station 6 were taken from this bed.

tamination during November. The last two samples were taken from the general area of the control stations. The oysters of all samples were taken from cultivated beds except those from Sugar House Bayou, which were wild growth ("coon" oysters).

These samples showed a rather high winter incidence of the disease and moderately high weighted incidence. All samples showed that a small percentage of oysters was in advanced stage of disease, indicating that even in winter deaths were occurring. Past studies have shown that the infections of late summer and fall are usually carried to mid-winter or later. The month of March normally will show a decrease in intensity of disease over the preceding January (Mackin, 1956). This seems to be the case in these studies. General surveys of the stations for level of incidence and intensity of *D. marinum* in live oysters were not taken in March, but were taken from April to September. The April incidences and intensities show a general decrease from the levels shown in pre-study samples of January and February (Table 5).

Measurement of disease intensity at the seven stations from March to September. Seventy percent of all deaths of oysters were shown to be due to *Dermocystidium marinum* infection. Table 4 shows that the mean of the stations was 71 percent. This figure includes Station 6, which developed a heavy early mortality from a disease other than that caused by *D. marinum;* the figure rises to 77 percent when this station is removed from the calculations. Station 2 and probably others also were affected by the new disease, but not to the extent as was Station 6. Table 4 also presents the data on recovery of gapers at all of the stations, the numbers of these destroyed by *D. marinum,* and the percentages of the whole. When the data are grouped for the control stations and compared with those from the experimental stations, in the former, 66 percent of the oysters died of *D. marinum* infection as against 74 percent for the latter.

TABLE 4

Numbers of oysters dying at the seven stations, number of gapers recovered, and number found to be in advanced stages of disease caused by *D. marinum*. Data collected in the period from March 3, 1957, to September 25, 1957

Station	1	2	3	4	5	6	7	Totals and means
Total dead in the period (gapers and boxes)	520	493	497	258	387	632	510	3297
Gapers recovered	224	138	105	115	142	202	162	1088
Percent recovery of dead oysters	43	28	21	45	37	32	32	34
Number gapers dying of *D. marinum* infection	184	90	77	77	108	82	145	763
Percent gapers dying of *D. marinum* infection	82	65	73	67	76	41	90	71
Total oysters dying of *D. marinum* infection	426	320	363	173	294	259	459	2294
Number of times checked	22	21	21	81	25	25	25	220

TABLE 5

Oyster death rates, case rates of disease caused by *Dermocystidium marinum*, and death rates resulting from this fungus disease of oysters, March to September, 1957. Figures are rates per thousand oysters per month

	March	April	May	June	July	August	September
			Station 1, Billet Bay				
Death rates, all causes	0.011	0.037	0.122	0.145	0.207	0.205	0.225
Case rates of *D. marinum*	....	0.607	0.617	0.662	0.812	0.911	0.867
Death rates, *D. marinum**	0.011	0.028	0.085	0.123	0.183	0.164	0.214
			Station 2, Rattlesnake Bayou				
	March	April	May	June	July	August	September
Death rates, all causes	0.011	0.047	0.145	0.174	0.221	0.131	0.092
Case rates of *D. marinum*		0.546	0.492	0.557	0.605	0.824	0.782
Death rates, *D. marinum*	0.004	0.046	0.103	0.098	0.158	0.075	0.069
			Station 3, Grande Ecaille				
	March	April	May	June	July	August	September
Death rates, all causes	0.014	0.039	0.125	0.131	0.202	0.187	0.171
Case rates of *D. marinum*	.	0.538	0.636	0.775	0.798	0.838	0.900
Death rates, *D. marinum*	0.000	0.028	0.080	0.098	0.152	0.166	0.171
			Station 4, Dredged Cut				
	March	April	May	June	July	August	September
Death rates, all causes	0.029	0.019	0.056	0.047	0.104	0.070	0.047
Case rates of *D. marinum*	. . .	0.399	0.607	0.554	0.578	0.683	0.764
Death rates, *D. marinum*	0.005	0.009	0.030	0.036	0.076	0.064	0.041
			Station 5, Sugar House Bend				
	March	April	May	June	July	August	September
Death rates, all causes	0.018	0.015	0.057	0.089	0.136	0.206	0.088
Case rates of *D. marinum*	....	0.622	0.469	0.647	0.730	0.884	0.944
Death rates, *D. marinum*	0.009	0.010	0.030	0.068	0.102	0.185	0.088
			Station 6, Sugar House Bend				
	March	April	May	June	July	August	September
Death rates, all causes	0.009	0.039	0.210	0.062	0.251	0.481	0.300
Case rates of *D. marinum*	.....	0.081	0.111	0.181	0.807	0.989	1.000
Death rates, *D. marinum*	0.000	0.000	0.010	0.023	0.213	0.470	0.300
			Station 7, Sugar House Bayou				
	March	April	May	June	July	August	September
Death rates, all causes	0.014	0.047	0.119	0.109	0.126	0.339	0.141
Case rates of *D. marinum*		0.690	0.626	0.821	0.911	0.948	0.965
Death rates, *D. marinum*	0.007	0.038	0.105	0.098	0.116	0.328	.....†

* Death was assumed to have been caused by *D. marinum* when the infection intensity was greater than moderate. Compare weighted incidences in gapers and in live oysters in Table 6.

† Data insufficient.

Figures 6 to 12 show in graphic form the monthly mortalities in terms of number dying per thousand population and on the same graphs are shown the relationship of the total deaths to deaths caused by *Dermocystidium marinum* infections alone. The

epidemic nature of this fungus disease is clearly shown in these graphs. Table 5 presents the data on total death rates, and death rates due to fungus disease. There are several points of interest to be derived from the data in this table. These are as follows:

(1) In April the mean number of deaths due to *D. marinum* per 1000 population was only five oysters. In July it was 143 and in August it was 208. These figures are means for all stations. The average increase in mortality from fungus disease from April to July was 2860 percent and from April to August it was 4160 percent.

(2) The number of deaths per 1000 cases of fungus disease was relatively low in April, averaging 23 oysters. In August the susceptibility to disease was much greater, when 207 out of each 1000 oysters infected died. This is one death per 4.8 cases. The greater susceptibility in August is probably due to increased reproductive activity of the fungus cells under stimulus of high temperature. Mackin (1953) reported experimental evidence that low temperatures retard development of the fungus. Owen (1955) also reported extensive experimentation showing the same effect and Hewatt and Andrews (1956) were able to prevent infection or development of infection in oysters kept at 15°C while those kept at 28°C died at a rate of 99 percent in six weeks.

(3) At the height of the epidemic at each station, deaths due to *D. marinum* far outnumbered deaths from all other causes combined. This was true at most other periods also but there were exceptional cases. Most notable of these was the early spring mortality at Station 6 which will be discussed in more detail later.

In Table 6 are listed the weighted incidences of disease caused by *D. marinum* in gapers, live oysters, and the total population. The weighted incidences of the total population were calculated from the weighted incidences of gapers and live oysters combined. For an explanation of "weighted incidence" see footnote under Table 6. In March the reliability of the data on weighted incidences in gapers is questionable because mortality rates were low and the number of gapers recovered for study was small. Data for succeeding months are considered to be reliable since the percentage of recovery was high (Table 4).

A study of Table 6 shows that there was an increasing weighted incidence of disease from April to the latter part of the summer. This increase is paralleled by the increase in total death rates and death rates due to disease caused by *D. marinum* as shown in Table 5.

At Station 6 the development of fungus disease is very interesting. At this station the oysters were approximately free of disease when placed at the station (Table 6). No deaths were recorded in March and April attributable to infection by *D. marinum* (Table 5). In May the death rate attributable to this disease was only 10 in 1000 oysters, and only 23 in June. In July case rates sky-rocketed and death rates increased sharply to more than 200 per 1000 population. In August the death rate reached a high of 470 per 1000 and 98 percent of the deaths were caused by the fungus. This phenomenal epidemic should be compared with that taking place at Station 5, remembering that Station 5 was only about 20 to 25 feet from Station 6. At Station 5, case rates in April were nearly 8 times as great as at Station 6, but the death rate from the disease reached only 185 per 1000 population in August. This is high, but does not compare with the 470 per thousand dying at Station 6. One might conclude that subjection of oysters to crude petroleum in some manner reduces death rate to *D. marinum* were it not for the

TABLE 6

Weighted incidence of disease caused by *D. marinum* in oysters at the seven stations. For explanation of the "weighted incidence" method for measuring intensity of disease in a population of oysters see explanation in the footnote* below this table

Period†	No. oysters at beginning of the period	Weighted incidence in gapers	Weighted incidence in live oysters	Weighted incidence of total population
		Station 1, Billet Bay		
March	1000‡	4.00		
April	989	3.94	0.71	0.83
May	892	3.38	0.66	0.99
June	723	4.02	0.91	1.36
July	568	4.51	1.27	1.92
August	404	3.90	1.95	2.35
September	271	4.80	1.32	2.10
		Station 2, Rattlesnake Bayou		
March	1000	2.50	.....	.....
April	989	4.50	0.68	0.86
May	883	3.41	0.78	1.16
June	695	3.00	0.76	1.15
July	524	3.75	0.83	1.48
August	358	3.00	1.18	1.42
September	261	4.25	0.78	1.10
		Station 3, Grande Ecaille		
March	1000	0.25	.....	.....
April	986	3.875	0.69	0.81
May	888	3.36	0.77	1.09
June	717	3.75	0.94	1.31
July	573	4.19	1.32	1.90
August	407	4.27	1.69	2.17
September	281	5.00	0.92	1.62
		Station 4, Dredged Cut		
March	1000	1.21	.....	.....
April	971	2.50	0.69	0.71
May	893	2.88	0.90	1.05
June	783	3.88	0.71	0.83
July	696	3.06	0.89	1.18
August	574	4.20	1.22	1.40
September	484	4.19	0.87	1.80
		Station 5, Sugar House Bend		
March	1000	2.00	.....	.....
April	982	3.17	0.89	0.92
May	907	3.00	0.69	0.82
June	795	3.76	1.01	1.26
July	664	3.65	1.50	1.79
August	524	4.50	1.83	2.38
September	376	4.50	1.82	2.06
		Station 6, Sugar House Bend		
March	1000	0.00	.....	.....
April	991	0.26	0.06	0.07
May	892	0.31	0.10	0.14
June	645	2.05	0.26	0.37
July	555	4.20	1.62	2.27
August	366	4.96	2.41	3.64
September	140	5.00	2.82	3.58
		Station 7, Sugar House Bayou		
March	1000	2.00	.....	.....
April	986	3.93	0.90	1.04
May	880	4.38	1.04	1.44
June	715	4.62	1.50	1.84
July	587	4.67	1.97	2.31
August	463	4.84	1.41	2.57
September	256	1.50	1.78	1.74

* Weighted incidence combines incidence and intensity of disease to make a convenient scoring device. When a sample of oysters is checked for disease, it is scored as follows: those oysters negative=0, those very lightly infected=0.5, light infections=1.0, light to moderate infections=2.0, moderate=3.0, moderate to heavy=4.0, heavy=5.0. The sum of the scores is divided by the number of oysters checked to give the weighted incidence.

† Because checks of the stations were not exactly on the first day or last day of each month, the monthly periods are only approximately correct. March and September were short months.

‡ Reduction in numbers of oysters shown in this column were not all due to deaths. Live oysters were withdrawn monthly for condition studies and for disease analysis.

fact that there were other differences between the two populations of oysters. The basic differences probably lie in two factors. These are as follows:

(1) Since the oysters at Station 6 had not been subjected to wholesale infection by *D. marinum* prior to removal to lower Barataria Bay, the population was composed of a very high percentage of susceptibles. The mortality process, weeding out susceptibles, had been going on for some time in Station 5 oysters, shown by the high incidence of *D. marinum* in the sample taken in January (Table 3). This high rate of infection must have been a carry-over from the preceding fall and summer. Hence, the percentage of oysters highly susceptible to *D. marinum* must have been considerably lower at Station 5 (and all other stations) than was the case at Station 6. Maxcy (1948) presented an excellent discussion of the effect of numbers of susceptible individuals in the development of epidemic disease in a community. He showed that the number of susceptibles in a population largely governs the development of epidemic waves.

(2) Immune reactions had already raised the capacity to resist development of severe disease in a high percentage of the oysters at Station 5. Over half of them were shown to be carrying infections in the winter and early spring, and additional oysters must have had the disease in the preceding summer and fall. Thus not only had the highly susceptible fraction of the population been eliminated by deaths, but, in a part of the remainder at least, individual immunity was increased through contact.

At some stations the last month showed a decrease in case rates and death rates to fungus disease. This is believed to be due to elimination of large numbers of susceptibles during the preceding high mortality periods. At some stations the peak of the summer epidemic was not reached until September (Stations 1, 3, and 7), and might have gone higher in October if observations had been continued.

Hewatt and Andrews (1956) also found that oysters from an endemic area had a greater resistance to infection and a sharply lower death rate when artificially infected than did oysters from a non-endemic area, *i.e.*, those which had not previously been exposed to the disease.

The high incidences shown in Tables 5 and 6 strongly suggest that all adult oysters in endemic areas in Louisiana become infected at some time during each summer. Infection and recovery from infection must be a constant process. The mechanism for throwing off an infection is a combination of phagocytosis and subsequent diapedesis of the phagocytes. Phagocytosis is a universal phenomenon in animals above unicells. Diapedesis probably is peculiar to molluscs. In the process, the phagocytes, carrying ingested foreign bodies, leave the oyster and are shed into the surrounding water. Orton (1924) observed diapedesis in diseased English oysters when studying the great mortalities of 1920–21.

New disease. An examination of the graph, Fig. 11, shows that at Station 6 (master control) there was a mortality of oysters which could not be ascribed to *D. marinum* infection. Neither was it caused by predators (conchs, etc.). This mortality of oysters began in April, reached a peak in May and subsided in June.

Gapers from this station which died in the period of this "unusual" mortality were sectioned for microscopic study. The stained slides of the tissues show that a disease was present in all these oysters, with one or two exceptions. The disease differs from that caused by *D. marinum*, as indicated by the histopathology. It has appeared in our studies before, but never in such form that the epidemic could be clearly defined and measured in extent.

At Station 2, some of the oysters which died in May and June were also victims of this new disease. Efforts are being made to determine what is the causative agent but no data will be presented here.

Miscellaneous contributors to mortality. Other than disease, most mortality of oysters is attributable to crabs, particularly to stone crabs *(Menippe mercenaria)*. Mackin (1947) and Menzel and Hopkins (1954) presented considerable data to show that the stone crab is highly lethal to oysters. Since stone crabs entering the trays were small compared with those found on oyster beds, they probably were not as effective as those used in experimentation by the authors cited above. *Callinectes*, the blue crab, has been said to be effective in destruction of oysters (Lunz, 1947). This author lists the blue crab as the most important enemy of oysters in ponds in South Carolina. Experiments in Louisiana are not convincing that *Callinectes* destroys large oysters, but this crab, like all others, probably takes a small toll.

The conch *(Thais floridana)* undoubtedly killed some oysters, especially at Station 3 where large conchs were present in numbers throughout the warm months. The large conchs can kill oysters without entering the trays. Adult conchs were not numerous at other stations and were almost completely absent at Station 7. The presence of immature conchs at Stations 5 and 6, where adult conchs were absent, will be discussed in a later section.

Some gapers showed the familiar muscle abscess resulting from penetrations of the shell under the adductor muscle by *Polydora*. Some deaths probably resulted from this abscessed condition. This peculiar damage has been described by Mackin and Cauthron (1953). These authors also gave a review of the literature on mudworms as a cause of disease in oysters. Several authors cited by Mackin and Cauthron (1953), believe that *Polydora* is a cause of extensive mortalities, but the point has not been satisfactorily demonstrated. That the worms cause small to moderate mortality seems to be well established.

Owen (1957) found a positive correlation of intensity of infestation of oysters with *Polydora* with high mortality areas in Louisiana. He set up certain experiments to check on the field results, and concluded from these that *Polydora* was not a "direct killer", but that the worms had a weakening effect, retarding growth. He also found that "*Polydora* is a part, like *Cliona, Thais, Martesia, Menippe, Callinectes* and others that is positively correlated to high mortality of oysters, . . ." The extent of the damage is still subject to question.

Recently, Loosanoff (1956) and Hopkins (1956) have called attention to *Odostomia*, a small snail parasite of oysters. This snail is so small that it is easily overlooked in field studies, even when it is present in large numbers. Loosanoff thinks that *Odostomia* probably cannot kill large oysters, but that it interferes with normal development and growth. Hopkins found that *O. impressa* attacks large oysters. The extent of damage to the oyster by this ectoparasite has not been experimentally measured. It may be a more important factor than it is now given credit for. It is reported as present at three of the stations in this investigation but apparently was not very numerous anywhere. Since it is so easily overlooked it may have been present in larger numbers than is indicated in Table 12.

Smothering with mud. At Station 3, the field data recorded that mud was found in the trays on May 23, June 13, and July 2. On the latter two dates several of the dead oysters were covered with the mud, and the thioglycollate records show that on these

dates some of the gapers were not killed by *D. marinum*. It is assumed that some of these died of smothering. Accumulated pseudofaecal and faecal material, *Polydora*, and *Corophium* seem to be the factors responsible for the mud.

Growth of Oysters at the Various Stations

On January 23, 1957, a sample of oysters was taken from the dredge-cut bed of the Freeport Sulphur Company. The oysters of this sample were carefully measured for shell length to establish the size range, modes and mean sizes of oysters used in this study. The graph, Figure 5, shows the size distribution of the oysters in the sample. Sizes used to stock the stations (excepting Station 6) included those over 30 mm in length. One group of 100 oysters at each station ranged from 30 to 50 mm in length and were referred to as "small"; the "large" group measured above 50 mm and numbered 200 at each station (two to nearly three inches). In mortality checks the "small" oysters were not separated from the "mortality, large" group, since the difference in death rates proved not to be great. The two groups were kept separate so far as growth data were concerned and the growth increments were calculated separately.

Growth of shell was calculated from January 23, 1957, when measurements were made, although the stocking of the stations took place during the following month. There were probably small differences in mean size of the oysters placed at the different stations but they probably were not significant.

Table 7

Shell growth of oysters used in these studies. Two different size groups were placed at each station. Oysters of the two groups had the following mean sizes on January 23, 1957; large, 67.2 mm (slightly more than 2½ inches); small, 38.7 mm (slightly more than 1½ inches)

Station	Group: small or large	Size on September 3–4, 1957	Increment mm	Increase in shell length (percent)
1	S	64.8	26.1	67
	L	94.6	27.4	41
2	S	61.4	22.7	59
	L	90.1	22.9	34
3	S	59.6	20.9	54
	L	87.1	19.9	30
4	S	62.9	24.2	63
	L	81.9	14.7	22
5	S	70.1	31.4	81
Control	L	91.3	24.1	36
7	S	64.2	25.5	66
Control	L	94.8	27.6	41

Table 7 summarizes the data on shell growth. Station 1 (experimental station) and Station 7 (control) had almost identical records. Station 4 had the poorest record so far as the large oysters were concerned, coupled with the least mortality. One might think that there was a connection were it not for the fact that the small oysters at the same station grew about as well as those at Station 7, which shared with Station 1 the record for best growth. The reason for the discrepancy in growth rates of small and large oysters at Station 4 is not known.

Growth at all stations was rapid and considerably better than for most oysters. It should be remembered that the growth records are based on survivors only. Comparison

of these rates with those observed by other authors is interesting. Ingle and Dawson (1952) made an extensive study of growth of oysters in Apalachicola Bay, Florida. They reviewed a considerable number of studies of growth of *Crassostrea virginica* made by other workers and concluded that "growth of the American oyster in Apalachicola Bay is more rapid than in any other region of its range that has so far been studied". They used shell length as a criterion of growth, and studied a number of populations of oysters of different sizes. They summarized their data with a graph, which showed the rate of growth per week for oysters of different sizes. This graph shows that oysters of the size of our "small" group grew at a rate of 1.1 mm per week. Our period of study was about 32 weeks, so the increment in oysters of this size class in Apalachicola Bay was about 35 mm. This is better than our best record for oysters of this size, but not too much better. Oysters of the size of our "large" group, using the same graph, would grow about 19 mm in Apalachicola Bay. This is less than the growth at any of our stations excepting Station 4. The growth of both large and small oysters at all stations had a mean of 24 mm, while in Apalachicola Bay it was 27 mm. At two of the stations (1 and 7) the mean growth was equal to the Apalachicola Bay growth mean.

If Ingle and Dawson are correct in their belief that growth of oysters in Apalachicola Bay is the best in the range of the species, then growth at our stations was next to the best and better than any other rate recorded other than at Apalachicola Bay.

Mortality seems to be high at all stations, but in terms of production or yield, which balances growth against mortality, the record was good at all stations. The lowest yield per sack planted was at Station 3, where the yield was one for one. At all other stations the volume of oysters increased. The data are presented in Table 8.

TABLE 8

Yield of oysters in sacks per sack planted. Calculated number of sacks* planted at each station: large oysters 0.251; small oysters 0.025. Mortality tray only; 200 large oysters and 100 small ones at each station

Station	Group	No. of survivors	Mean size mm	Approx. no. per sack†	Volume in sacks	Number of sacks per sack planted	Means
1	Large	89	94.6	280	0.318	1.26	1.27
	Small	29	64.8	900	0.032	1.28	
2	Large	100	90.1	340	0.294	1.17	1.42
	Small	42	61.4	1000	0.042	1.68	
3	Large	72	87.1	380	0.189	0.76	1.00
	Small	34	59.6	1100	0.031	1.24	
4	Large	144	81.9	460	0.312	1.24	2.12
	Small	72	62.9	960	0.075	3.00	
5	Large	121	91.3	340	0.356	1.42	2.22
	Small	53	70.1	700	0.076	3.03	
7	Large	87	94.8	280	0.310	1.24	1.68
	Small	48	64.2	900	0.053	2.12	

* A sack of oysters is equal to 0.053 cubic meters.
† Number per sack in each size from Hopkins (1950), Fig. 1.

Set and Growth of Young Oysters

All stations had accruals of spat during the studies. When oysters were placed in the trays, all young oysters which could be seen were removed from the shells. Most oysters were culled and placed at the stations in February. During February a set of oysters occurred following a period of mild temperature in mid-winter. Water temperatures during the last four days in January were 20° to 22°C and in February the first 12 days had temperatures between 22° and 24°C. The small oysters attached to the adult oysters in the latter part of February, and were mostly too small to be seen and removed in the original culling operation.

Later sets occurred in April and May. The first week in May produced a general spawning. Thereafter, through the summer, continual spawning at a low rate was the rule. Thus the oysters in the trays had a more or less continual accrual from the time they were placed in the trays until September, excepting only parts of March and April.

Because boxes were removed all set counted was on the live oysters and on the trays. In early March, wood partitions were placed in some of the trays. These picked up spat almost as soon as placed in the water. The maximum size of these spat in early September gives a good idea of growth for the six months of the study. The data are contained in Table 9.

Table 9

Maximum size of spat attached (1) to partitions in trays placed in the water in early March, 1957, and measured September 3–4, 1957, and (2) to oysters, which attached in early February

Station	(1) On wood partition, March to September size of largest spat mm	(2) On adult oysters February to September mm
1	no data	43
2	41	42
3	43	54
4	no data	68
5	no data	64
6	40	50
7	55	40

These young oysters grew about 1½ to 2½ inches during a six to eight month period. This represents maximum growth for the area. Young oysters, unlike those more than a year old, grow in summer as well as in winter in Louisiana. Older oysters often show a net loss of shell length in summer, due to cessation of growth and crumbling of the thin shell edge.

Numbers of Young Oysters Setting at the Different Stations

Counts were made of the numbers of young oysters found attached to the older oysters at the end of the study. These all attached between February and September. The numbers were calculated in terms of number per 10 oysters in the trays. The data are contained in Table 10.

Best setting occurred at Station 2. Set was so heavy and growth of the young oysters so rapid that the older oysters were well covered with the new year class of 1957. Station 7, a control station, had the next best record, and Station 4 the poorest.

TABLE 10

Numbers of spat attached to oysters at the end of the study (September 3–4). Numbers are calculated per 10 oysters

Station	1	2	3	4	5	6	7
No. per 10 oysters	17.9	94.5	13.0	2.3	21.5	25.7	57.5

INDEX OF CONDITION

Index of condition is a measure of the "fatness" of oysters, expressed by the ratio: dry weight of the meats in grams ÷ shell capacity in cc × 100. Sometimes called the "coefficient of fitness", it may not, in fact, reflect either condition or fitness. The name implies that when the index is high that the oyster is healthy or physically fit, which may or may not be true, just as a fat man may be neither healthy nor fit. Ingle and Dawson (1952) remarked on the fact that growth of their oysters in Apalachicola Bay, Florida, was very rapid while they were relatively low in glycogen, and certainly rapid growth is a primary criterion of physical fitness. It seems better to consider the index of condition as an index of marketability rather than of condition in the sense of physical vigor with which it is usually associated.

TABLE 11

Condition of oysters at the different stations from April, 1957 to September, 1957 Volumes in cc; weights in grams

Station		April	May	June	July	August	September
1	Mean shell capacity	27.60	28.00	40.50	24.00	20.00	23.00
	Mean dry weight	2.87	2.02	1.98	1.32	1.84	1.13
	Index of condition	10.4	7.2	4.9	5.5	9.2	4.9
2	Mean shell capacity	26.95	22.80	22.50	20.30	18.50	19.00
	Mean dry weight	2.23	1.32	1.22	1.23	0.73	1.09
	Index of condition	8.3	5.8	5.4	6.1	3.9	5.7
3	Mean shell capacity	21.15	29.20	32.00	24.50	23.70	28.50
	Mean dry weight	1.80	2.68	1.89	1.56	0.95	1.25
	Index of condition	8.5	9.2	5.9	6.4	4.0	4.4
4	Mean shell capacity	26.75	24.00	22.50	29.00	22.20	26.50
	Mean dry weight	2.64	1.63	1.05	1.39	1.14	1.22
	Index of condition	9.9	6.8	4.7	4.8	5.1	4.6
5	Mean shell capacity	29.85	26.70	29.40	21.70	26.50	29.50
	Mean dry weight	3.50	2.90	1.92	1.57	1.59	1.54
	Index of condition	11.7	10.9	6.5	7.2	6.0	5.2
6	Mean shell capacity	24.60	22.50	23.70	27.50	27.00	24.50
	Mean dry weight	2.13	1.70	1.45	1.57	1.78	2.45
	Index of condition	8.7	7.6	6.1	5.7	6.6	10.0
7	Mean shell capacity	24.50	33.00	21.80	30.00	25.20	29.70
	Mean dry weight	2.45	2.49	0.95	1.32	1.20	2.37
	Index of condition	10.0	7.6	4.4	4.4	4.8	8.0

The data from the study of "condition" are presented in Table 11. As was to be expected there was a general drop in condition during spawning in April and May, after which the oysters just held their own during the summer. Control stations had a markedly better record in the last part of the summer than did the experimental stations, (especially at Station 1) in August, while spawning was only moderate at the control stations. Altogether, differences between the experimental stations and control stations were not significant. The range of values between two control stations was

greater than the range between the means of the experimental and control stations in every month of the test.

Animal Communities of the Stations

The principle that detrimental pollution of water bodies can be detected by analysis of the animal community existing in an area is well established. A normal community exists where conditions are normal; where conditions are abnormal, varying degrees of alteration of the community takes place. Efforts at standardization of procedures in analysis of pollution effects have been made (Roberts, 1949). This and nearly all other efforts have dealt primarily with the problems of stream pollution. The principles apply irrespective of the type of water body involved. Extreme modification of an environment eliminates all but a few species of organisms, and of the few remaining, one or more may develop enormous populations, overshadowing all others. Gradations from the extreme condition may be detected, resulting in less pronounced modification of the community.

The use of the community in judging the normality of the environment depends on the establishment of the composition of the normal community, the ability to detect omissions of significance, and the recognition of populations of exotics which are stimulated to growth by abnormal concentrations of toxic substances.

It is nearly always assumed that a foreign substance introduced into a water body is harmful. It is not always so. The establishment of the presence in an area of a normal community may only mean that a foreign substance is not toxic and is incapable of so modifying the habitat as to produce an effect.

Establishment of the composition of the normal community. Gunter (1955) has listed the common animal associates of oysters at stations in Texas bays having relatively high salinity. He noted the presence of: *Menippe, Thais, Cliona, Microciona, Hydroids,* Ctenophores, *Aurelia,* and Bryozoa.

Compared with our list, Gunter's is quite short, but evidently contains the same basic community elements.

Hedgpeth (1953) discussed what he termed the "oyster biocenosis" of the Gulf of Mexico. The macroscopic invertebrate animals mentioned by him are listed below: *Stylochus, Callinectes, Menippe, Odostomia, Cliona, Polydora, Bugula, Thais, Brachidontes* and *Crepidula.*

Mackin and Wray (1950) listed a larger group of invertebrates and small fishes observed at Sugar House Bend (the control location). The observations were spread over a 14-month period in 1948 and 1949. This list was as follows: *Cliona celata, C. truitti, Haliclona sp., Aiptasia pallida, Campanularia sp., Astrangia astreiformis, Stylochus ellipticus, Membranipora sp., Bugula neritina, Nereis limbata, Polydora websteri, Martesia smithi, Brachidontes recurvus, Congeria leucophaeta, Crepidula plana, Anachis avara, Corambe sp., Thais floridana, Balanus eburneus, Petrolisthes armatus, Menippe mercenaria, Panopeus herbstii, Clibanarius vittatus, Callinectes sapidus, Eurypanopeus depressus, Crangon armillatus, E. dissimilis, Palaemonetes vulgaris, P. carolinus, Corophium lacustre, C. louisianae, Molgula manhattensis, Gobiesox virgatulus, Gobiesoma bosci, Hypsoblennius ianthus, Chasmodes saburrae,* and *Opsanus tau.* This list should be compared with that compiled in the present study (Table 12).

TABLE 12

Summary of the groups and genera of macroscopic animals found at the stations

Genus or group	Station					
	1	2	3	5	6	7
Cliona celata	(x)	(x)	(x)	(x)	x	(x)
Haliclona sp.	(x)	(x)	(x)	(x)	(x)	(x)
Aiptasia pallida	x	–	x	x	x	x
Membranipora sp.	(x)	(x)	(x)	(x)	(x)	(x)
Stylochus ellipticus	–	x	x	–	–	–
Polydora websteri	(x)	(x)	(x)	(x)	(x)	(x)
Martesia smithi	x	x	x	x	x	x
Brachidontes recurvus	(x)	(x)	(x)	(x)	(x)	(x)
*Bankia sp.**	(x)	(x)	(x)	(x)	(x)	(x)
Corambe sp.	x	–	–	–	x	–
Thais floridana	x	x	(x)	(x)	(x)	x
Odostomia impressa	x	–	x	x	–	–
Anachis avara	–	–	–	x	–	–
Crepidula plana	x	x	x	x	(x)	x
Balanus eburneus	(x)	(x)	(x)	(x)	(x)	(x)
Corophium lacustre	x	x	x	(x)	(x)	x
Isopoda	–	x	–	x	–	–
Crangon armillatus	x	(x)	x	x	x	–
Palaemonetes vulgaris	x	(x)	x	x	(x)	(x)
Menippe mercenaria	x	x	x	x	x	x
Callinectes sapidus	(x)	(x)	(x)	(x)	(x)	(x)
Eurypanopeus depressus	(x)	(x)	(x)	(x)	(x)	(x)
Panopeus herbstii	–	x	x	x	x	x
Petrolisthes armatus	(x)	(x)	(x)	(x)	(x)	–
Clibanarius vittatus	x	x	(x)	(x)	(x)	x
Molgula manhattensis	–	–	–	x	x	–
Gobiesox virgatulus	x	x	x	x	x	–
Gobiosoma bosci	(x)	(x)	(x)	(x)	(x)	x
Hypsoblennius ionthas	x	(x)	(x)	(x)	(x)	x
Chasmodes saburrae	–	x	x	x	x	–
Opsanus tau	x	x	x	x	x	–
Fundulus similis	–	(x)	–	(x)	x	–
Lagodon rhomboides	–	x	–	x	x	x
Chaetodipterus faber	–	–	–	–	–	x

* Records do not show the presence of shipworm until the end of the study, when it was observed in numbers in the board partitions of the trays. It is certain that it was present earlier, but just when the penetration of boards began is not known.

The lists presented above indicate that the basic elements of the faunal complex are nearly the same over wide estuarine areas of the northern Gulf. Faunal elements expected to appear in the normal community may be judged from these lists.

Macroscopic invertebrates observed at the seven stations in this study. On January 23, 1957, the planting by the Freeport Sulphur Company in the dredged cut was examined and a short check of the fauna was made. At this time oil could be stirred from the bottom but the oysters did not appear to be oily. The fauna of the bed appeared normal. The following animals were observed: *Cliona celata* (relatively heavy infestations in some oysters), *Martesia smithii* (light infestations of the shell), *Crepidula plana* (very numerous), *Molgula manhattensis* (abundant), *Polydora websteri* (abundant), *Eurypanopeus depressus* (abundant), *Callinectes sapidus, Membranipora sp.* (abundant), *Petrolisthes armatus, Haliclona sp.* (few shells covered), *Balanus eburneus* (many), *Stylochus ellipticus, Brachidontes recurvus* (many attached to oysters), and *Odostomia impressa.*

Although the check was hurried, it established that the usual community was present at that time.

Table 12 presents the list of macroscopic animals found in this study at the various stations. Where species were easily recognizable, specific names have been given. In others, generic names have been given. In some genera, and in some major groups, more than one genus or species was present.

In the table an "x" means that the organism was present; if the symbol (x) is given, the genus or group was abundant in at least one period of the study. Some of the animals were constantly present from early March to September, when the study ended. Table 13 summarizes the data for the various stations.

TABLE 13

Number of genera of macroscopic animals observed at the various stations

Station	1	2	3	5	6	7
Number observed	29	33	30	35	34	26
Number becoming numerous during the study	15	16	18	21	21	14
Number continuously present	10	11	10	10	6	7

Station 4 is not represented in the table. At that station data on the fauna were taken at widely spaced intervals, but not consistently, and it was thought best to omit the station from this part of the study altogether. However, all of the common forms, fifteen in all, were recorded there.

There are some observations on certain species which are worth recording. When the oysters used at all stations (excepting 6) were taken from the bed in the dredged cut, the oysters carried a certain epifauna with them to the stations. *Cliona* (boring sponge) was present in the shells of nearly all oysters and remained abundant at all stations to the end of the study. But those oysters used to supply Station 6 had no sponge, and picked up only a light infestation in a few of the oysters before September. This set up an experiment contrasting the mortality of oysters with and without severe sponge infestation. Station 5, adjacent to Station 6 and with an identical environment, was stocked with oysters all of which were more or less heavily infested by boring sponge growth in the shell. Mortality at this station was materially less than that at Station 6 where more than 90 percent of the oysters were free from infestation. While this circumstance cannot prove that sponge infestation never becomes so heavy as to injure oysters, it does show that in Louisiana waters the ordinary "heavy" infestations have no noticeable effect. There are numerous references in the literature, indicating that mortalities from shell-boring sponges occur in various parts of the world. In fact Dollfus (1922), discussed it as a disease of oysters, calling it the "maladie du pain d'epice". Oysters have been observed in South Carolina so heavily infested that penetrations through the shell, especially those entering the adductor muscle area, seemed quite capable of destroying the host oyster.

Thais floridana (conch) was common at Station 3 and was present in moderate numbers at all times. At other stations the appearance of the conchs was intermittent. At Station 1 they were never abundant and disappeared through most of July. At Station 5 they disappeared in May, returned briefly in June, disappeared again through July, to reappear in August. Adult conchs were not seen at this station or at Station 6 during the entire study although they were nearly adult size at the end of the study. Station 7 was free of conchs excepting a few which were seen on March 29.

There were abundant small hooked mussels attached to the oysters when the stations were stocked. These increased in number at all stations during the study, possibly because of relative scarcity of conchs. Conchs, when found in the trays, were always removed to prevent excess loss of oysters to this predator. However, it is believed that they would never have become an important factor, except at Station 3, because of the presence of large numbers of spat and mussels, which they destroy before attacking large oysters.

Table 13 condenses the data on numbers of genera or groups of genera found at the six stations.

Discussion and Conclusions

If all of the stations were equal in all respects excepting that the oysters were not subjected to crude oil contamination at Station 6, one would be compelled to believe that subjection of oysters to crude petroleum is beneficial so far as survival is concerned. That thesis cannot be accepted. The data clearly show that contact with oil over an extended period had no effect so far as survival is concerned. This conclusion is supported by the fact that at experimental Station 3 the mortality rate for the period of the study was 61.2 percent and 65.0 percent (see Table 2), and in the same period in 1949 it was 63 percent. Also the mortality rates at the four experimental stations approximated that predicted by the year-class analysis method.

Two kinds of controls also show that there was no effect of the crude oil on survival. Those oysters at Stations 5 and 7 had been subjected to oil but removed to an uncontaminated area where further subjection did not take place. The experimental oysters remained in the polluted area. The polluted area obviously did not remain that way for the duration of the study, except at Station 2. This latter station was intermittently subjected to light oil slicks for the duration of the study. There were, therefore, three grades of subjection to crude oil pollution, with the oysters at Station 2 getting the greatest dosage, other experimental stations less, and the two modified control stations still less. Station 6 was the master control. Under these conditions if oil contamination had anything to do with the mortalities that occurred, Station 2 oysters would have had the highest mortality rate, Station 6 the least and the others would have ranged in between. This did not occur.

The timing of the mortalities is also important in judging the effects of the oil. Minor mortality occurred in the period from November when the Mecom well was flowing oil, to April, while all significant mortality occurred in the summer from June to September. The general mortality pattern was the same irrespective of the location of the stations. This excepts the May mortality peak at Station 6.

Attention has been called to the opportunity presented in contrasting the development of fungus disease at Station 6 where oysters were not subjected to oil and at Station 5, located in the same area, and containing oysters which were subjected to oil in the early period of the study. Station 5 oysters had a considerable infection with fungus disease, while the oysters used to stock Station 6 had almost none. Station 2 should also be compared with Station 6. Examination of the data shows that subjection to crude oil pollution had no synergistic effect in increasing *D. marinum* infection or intensity of infection. The theory that oil may in some manner be prerequisite to development of disease is disproved in view of the fact that Station 6 oysters developed the highest

weighted incidence of disease ever observed in the field and a corresponding destructive epidemic.

The oysters at Station 2 were subjected to the full effects of all factors incident to drilling in an oil field which was being actively exploited. Survival of oysters, growth, set of spat, growth of young oysters, and development of a normal associated fauna were in no way inhibited at Station 2 as compared with other stations.

Dermocystidium marinum was shown to be the most important agency producing deaths at all stations. The form of the epidemics was typical of fungus disease in the past, responding to high summer temperatures with rapidly increasing death rates. There were other, more minor, agencies causing death of oysters.

The environments of the experimental stations were not modified by crude oil to the extent that any change in the normal oyster reef community occurred. Control and experimental communities did not differ significantly; no species normally present in Gulf oyster communities was absent at experimental stations. Since the oil was present in considerable amounts over a minimum period of two to three months the only conclusion possible is that accidental crude oil spillage in very large amount did not significantly modify such communities. Some of the animals constantly present at the stations had the capacity to leave (*i.e.*, fishes, crabs), but were quite evidently attracted to the trays. Normal reproduction and growth of populations took place during the entire period of the study.

The oysters themselves spawned normally, and heavy sets of young oysters occurred at some experimental stations. These young oysters grew rapidly with obviously small mortality, while at the same time the older oysters died of epidemic disease in large numbers. The difference in susceptibility of young oysters to fungus disease has been established by Ray (1954b) and Owen (1955). If crude oil had had anything to do with the mortalities, the young oysters and the older oysters would have died in approximately equal percentages.

Lastly, growth of the surviving oysters was excellent, as was their condition. Thus neither survival, reproduction, growth or size of the oyster meats was affected by the oil.

Persistence of Oily Taste

It has been established (Menzel, 1948) that losses of crude may result in production of oily taste. It has also been shown that oysters will readily ingest crude oil (Mackin, 1948). It was shown in the latter study that ingestion of crude oil in no way interfered with the feeding of oysters. The large amount of crude oil lost by the Mecom well unquestionably imparted an oily taste to some oysters in the areas of heavy concentration of oil. Since the Freeport Sulphur bed in the dredged cut was probably the most heavily contaminated of any in the area of the oil loss, it was proposed to check this one for disappearance of oily taste. However, on December 26, 1956, when the bed was visited by the authors and Mr. Fred Deiler, no certain oily taste could be detected, although the senior author checked one oyster which may have had a slight oil taste. This oyster was abnormal, but the usual kerosene taste was not present. All other oysters checked were negative on that date and subsequent checks, which were soon abandoned, showed no oily-tasting oysters. In this connection it should be realized that three "tasters" cannot test a large sample. Since, as recorded above, oil could be stirred from the bottom at this time, it is not certain that there were no "oily" oysters on the bed, bu the percentage

must have been very low. Oysters once rid of oily taste do not necessarily remain so indefinitely. If oil droplets are in the mud a high wind may put them into suspension to be ingested by the oysters. However, no recurrence was recorded. As oil is subjected to weathering, those fractions containing the aromatics responsible for oily taste are lost by evaporation, leaving the heavy oils which are largely tasteless.

Acknowledgments

Supervision of the study was the responsibility of the authors. Mr. James Boswell made all thioglycollate cultures for determination of the incidence and intensity of *Dermocystidium marinum* infections in the oysters. Mr. Boswell also made most of the readings of the cultures, *i.e.*, he prepared the requisite slides from the cultures and determined the incidence and intensity of disease. He also fixed the gapers for sectioning and did the actual sectioning. Staining of these slides was done by the senior author. Mr. Boswell made most of the determinations of condition, but was assisted in this by Mr. Vernon Thatcher. Mr. Thatcher who was in charge of the field work, was assisted by Mr. Ed Schrader and Mr. Patrick Landry, and occasionally by the authors. Field work consisted of regular visits to the stations, when all oysters were examined individually, gapers were collected, as well as samples of live oysters for thioglycollate cultures and condition analysis.

The field work at Station 4 was supervised by Mr. Fred Deiler of the Freeport Sulphur Company, and the actual checks at this station were by Mr. J. D. Bergeron. Ed Schrader made the field surveys for oil contamination, assisted by Mr. Patrick Landry.

The condensation of the data was the responsibility of the junior author, and the writing of the report and interpretation of the data were the responsibility of the senior author.

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Canal Dredging and Silting in Louisiana Bays[1]

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This paper was derived in part from *Canal Dredging and Silting in Louisiana,* a report from the Texas A & M Research Foundation Marine Laboratory (Mackin, 1960a).

Abstract

Effects of dredging on oysters were studied in shallow bays of Louisiana and discussed relative to the normally high turbidities of an eroding marsh area. Data were taken from unpublished reports of Projects 9 and 23 of Texas A & M Research Foundation.

High levels of turbidity and organic matter were measured in the bays of Louisiana and related to action of tides, waves, general land subsidence, disintegration of the shorelines, and seasonal patterns of equilibrium between scouring and redeposition. With detailed examples from the Lake Grande Ecaille area, erosion of the marsh islands was demonstrated as accentuated by the Mississippi levees, which no longer allow influx of river silt. Calculations were made on sediment load, settling velocity, and transport.

Three case histories of dredging were followed, one a hydraulic dredge depositing a spoil island, one a clam dredge operating in a canal, and one a hydraulic dredge depositing in a half-fan circle. Silt was carried a maximum of 1300 ft. from the dredge with rapid dispersal by dilution and sedimentation. A maximum of one percent of the material drifts away from the site of operation. Turbidities beyond a few hundred feet did not exceed the natural levels (20 to 200 ppm). Experiments showed no harmful effects on oysters of turbidities tested (5 to 700 ppm).

Experiments indicate that dead oysters did not consume oxygen during decomposition at rates much greater than live oysters. Oxygen demand of sediments dispersed in dredging was relatively less than the surface sediments. Oxygen depletion with dredging in Louisiana marshes was found to be a relatively minor factor not responsible for mortality.

Cores were compared microscopically from Lake Grande Ecaille in control situations, in areas receiving light silt from dredging, and in areas of heavy silt deposit in spoil deposition. Inferences were drawn from concentrations of foraminifera from winnowing, resuspension of fossils no longer living in Louisiana, and the presence of shell and silt material together. Many soft silt conditions affecting the distribution of oyster reefs were traced to marsh erosion, disintegration, and subsidence, with cores unmistakably different from those in spoil deposits. Some legal controversies were discussed.

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[1] Oceanography and Meterology Series from the Department of Oceanography and Meterology, and A & M College of Texas. A report from Project 23E, Texas A & M Research Foundation, sponsored by Texaco Incorporated, Humble Oil and Refining Company, The California Company, Gulf Refining Company, Phillips Petroleum Company, and Shell Oil Company.

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Introduction

The aim of this study is to examine the conditions in the Mississippi River Delta which have to do with natural and artificial silting and its relation to oysters. Silting as produced by canal dredging is artificial in that the dredges are not a part of the natural environment. But after silt is suspended in the water by artificial means, it behaves as it would if natural suspending agencies, the wind, waves, and flooding, had put it in suspension. From the time it is suspended to the time it comes permanently or semi-permanently to rest on the bottom, it is controlled by natural forces.

The effects of direct deposition of spoil on oysters or oyster beds are not considered here. Such effects are obvious. Most emphasis in this study has been placed on observations of water transport of silt put in suspension by dredges. The effect of canal dredging on oxygen levels was also examined. Field studies were concentrated in the estuaries of Lake Grande Ecaille, Louisiana (Fig. 1). The effect of suspended silt on oysters has been tested experimentally, and the results are included in this paper, extending the results of previous researches.

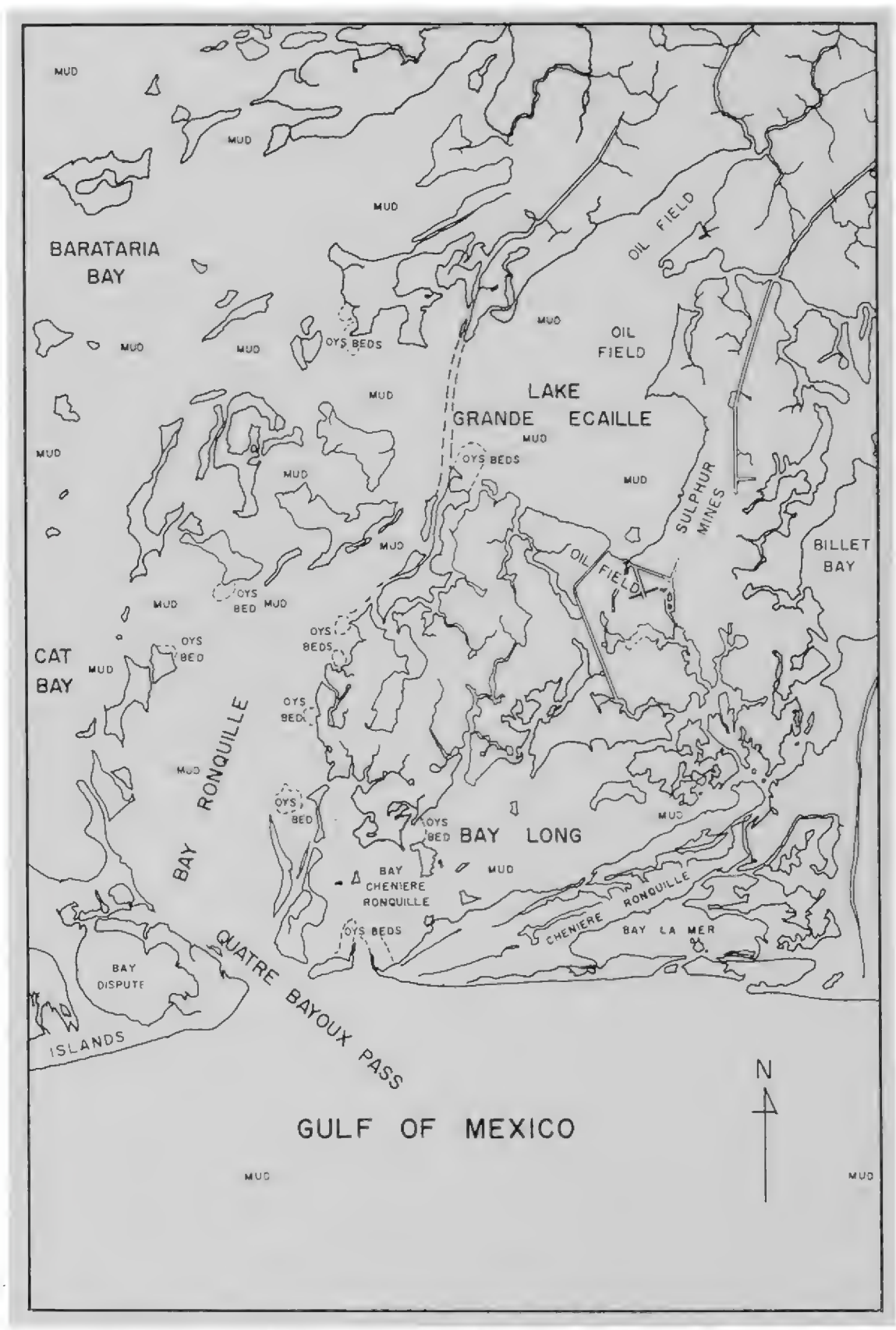

FIG. 1. Map of Lake Grande Ecaille, showing (1) old bayou crossing the west end, (2) transect areas, (3) general locations of oil fields and sulphur mine.

History and Literature

Cary (1906) noted that many canals were being dredged in the coastal marsh areas of Louisiana and stated that these canals were ruining the oyster industry. According to Cary, canals allowed large amounts of fresh water to enter the bays, lowering the salinity to a dangerous level during freshets. Apparently, the canals referred to were drainage projects, or perhaps some were for navigation purposes. This was the first recorded complaint that canal dredging in the marsh areas was harmful to oysters.

Although canal dredging in the coastal waters has been a more or less steady accompaniment of increased population from earliest times, it was not until much later that the complaints were revived (Gowanloch, 1947; McConnell, 1952a, 1952b). Gowanloch and McConnell's concept of the source of damage differed materially from that of Cary nearly fifty years earlier. Silting of oyster beds was stressed, it was claimed that increased salinity resulted, and the danger from diversion of currents was pointed out. The claim that increase in salinity occurred in certain areas as a result of canal dredging was further developed by St. Amant, Fairchild, Friedrichs, and Strawbridge (1956) and St. Amant, Friedrichs and Hajdu (1958). These authors believed that canal dredging was one of several causes of increase in salinity over the last 25 to 30 years but, like McConnell, presented no data supporting their conclusion. Gunter (1957a, 1957b) presented a contrary view. This author pointed out several advantages of dredging as did Viosca (1958) who listed return of nutrients to the water by suction dredges when bottom muds are overturned, and the apparent preference of shrimp for the highly turbid waters near shell dredgers.

The earliest study of the effect of dredging operations on oysters was apparently that of Lunz (1938). He investigated the dredging of the intracoastal waterway by the United States engineers in the vicinity of Charleston, South Carolina. Lunz measured silting quantitatively, studied mortality of oysters in close proximity to the dredge, and concluded that oysters would not be destroyed by silt from dredging unless they were buried in it. From this and other studies (Lunz, 1952; Lunz and Bowman, 1955), coupled with a study of flooding of the Santee River in South Carolina (Lunz, 1936) he concluded that high turbidity caused by dredges was not harmful to oysters, and that direct physical destruction of reefs or burial of oysters had to take place in order that oysters would be destroyed by such operations.

Wilson (1950) made an extensive study of the effects of shell dredging on oysters in Copano Bay, Texas. He also carried out some experiments, subjecting oysters in aquaria to very high concentrations of suspended silt, and covered oysters with sand, silt, and oily sludge to determine how long they would live when buried. Oysters buried under silt survived for from four to five days, and turbidity of 4000 parts per million was "detrimental" to oysters according to Wilson's findings. Turbidity of 4000 ppm may be found close to the outlet of a suction dredge. More important, Wilson found that "suspended material as those which are caused by the dredge are dissipated rapidly in open areas." He also found that a year of operation of a suction dredge in Copano Bay "did not place and maintain enough material in suspension to be detrimental to oysters while in the suspended state."

Like Lunz, Wilson found that if an oyster is covered completely with silt, it will die, hardly a debatable matter. Oyster beds within 300 yards of the dredge were subject to

silting, but only if they were on flat, low bottom. Those beds in shallower water, or raised above the bottom, were not subject to siltation unless the spoil was directly deposited on them. Even a year of operation in one bay failed to produce any kind of effect at distances greater than 300 yards and deposition of silt at that distance was not measurable.

Ingle (1952), Ingle and Dawson (1953), and Ingle, Ceurvels, and Leinecker (1955) studied turbidity and dredging in Florida bays, and in Mobile Bay, Alabama. The first paper cited was a study of the distribution of silt from a shell dredger, operating in Mobile Bay, Alabama, and of the effects of dredging in general. The author found that nutrient compounds are released from bottom muds by dredging and returned to use in the biological cycle. Fish exposed to high concentrations of suspended silt were killed, but the author pointed out that if not confined in the silt, fish would avoid it. It was also found that silt from a dredge is rapidly precipitated and that "harmful turbidities created by dredges are limited to a small area in the vicinity of the sludge discharge outlet." Oysters within 50 yards of an active shell dredger were unharmed. Ingle thought that 100 yards was a safe distance and set 300 yards as the greatest distance (in Mobile Bay) that fine materials would be carried by currents and considered these fine materials as not harmful to oysters. Ingle, Ceurvels, and Leinecker pointed out that the muds appeared to have no toxic properties, and hydrogen sulphide was not present in the turbid waters near the dredge investigated. Ingle and Dawson (1953) believed that oysters in Apalachicola Bay were benefited rather than harmed by fine suspensions of silt.

A careful study of these papers shows that most authors found some sort of measurement of turbidity to be possible, but described no method that accurately measured deposition of silts on the bottom. The factors that control deposition seemed to be little understood.

Dredges and Spoils

Dredges differ in their operation according to the type. In Louisiana marshes, the clam-shell dredge is most generally used; in bays, the hydraulic ("suction") dredge operation is more often encountered. Draglines and occasionally the hydraulic jets are used for special purposes.

The hydraulic dredge has a revolving cutter which loosens the bottom silt. The spoil is then sucked into a 10- or 12-inch diameter pipe and is pumped out at the free end. The advantage of the hydraulic dredge is that it can deposit spoil at a distance from the working dredge in several ways: (1) pumping to land (that is, the marsh), (2) depositing all spoil in one area to form spoil islands, (3) distributing in a ridge alongside the canal by pumping forcibly out of a short fixed length of pipe, and (4) spreading over the bottom by mounting the end of the exit pipe on a barge which is moved over a wide strip of bottom paralleling the barge. Other types of dredges must deposit spoil immediately alongside of the canal being dug; this produces undesirable spoil ridges in bays, but is satisfactory in marshland dredging.

Pumping to land has the advantage that the spoil is removed from the bay and produces no significant turbidity, but the disadvantages may outweigh the advantages. Marsh in most of the area is not stable enough to hold a great weight so that the spoil

sinks, breaking up the marsh. Semi-fluid mud under the marshes is squeezed out, usually into the nearest adjacent bay or bayou. Unless small containing levees are built, much spoil may flow back into the bays, defeating the purpose of placing spoil on land. Lastly, if any great distances are involved the cost of dredging and pumping to the marsh may be prohibitive. The method of pumping to the marsh is, therefore, useful only when the marsh is stable, retaining levees are built, or the desposit of spoil is far away from any waterway. It is difficult in any case to keep costs within reason.

Pumping spoil to form isolated spoil islands in a bay has some advantages but more disadvantages. If an area of relatively stable bottom at a reasonable distance from oyster beds, channels, etc., can be found, spoil disposal in this manner is satisfactory. But the problem of prohibitive costs is encountered when any great length of canal is to be made. A spoil island also becomes a hazard to navigation, however placed. Most of it is under water and troublesome even when the apex of the mound is properly marked.

It is believed that the method of spreading spoils over a band two or three hundred feet wide parallel to a canal is by far the best method of disposal from the standpoint of industry and oystermen. Spoil pipe need not be more than 300 to 400 feet long, which will not add materially to costs, and the spreading of spoil on the bottom in a layer not over six inches in depth has little or no effect. Bottom covered in this manner is always stiffer than before (because of concentrations of shell from the dredging). The weight of the spoil causes compaction of soft bottom so that the decrease in depth of the water is not significant.

The method of disposal which results in the formation of an underwater ridge parallel to the canal has objectionable features. A long spoil ridge may so change wind fetch that effective turbulence from waves may fail to keep a bottom clear and cause it to become a depositing area. Current rates may also be reduced so that silting may result.

It is rare that cutting canals through open bays produces change of current or deposition of silt if proper spoil disposal is made. However, canals which cut across major marsh waterways are a hazard because of the possibility that currents are deflected from original courses. Deposit of silt will occur at any time that effective reduction of current takes place. Occasionally a bayou may be deepened by access to a canal if current velocity is increased. Active bayous (as contrasted with dead bayous) should be protected by blocking intersecting canals at their junctions with bayous as soon as possible.

Methods for Determination of Turbidity

A Fischer Electrophotometer was used to measure turbidity. A portable gasoline-driven Onan generator was carried in boats to provide power for determinations in the field. Samples were taken with a Foerst tube, transferred to the electrophotometer cell, and read immediately. Prior to each reading, the electrophotometer was adjusted to read 100 percent transmission through distilled water.

Light transmission readings were calibrated in parts per million against a standard candle in a Jackson turbidimeter with standards of fuller's earth prepared by W. C. Curtin Company. Samples over 1000 ppm were established by dilution.

Photoelectric cells were also mounted in a pipe so as to record light transmission in continuously flowing water. They were also calibrated against fuller's earth standards.

Coring Methods

A coring method was developed to provide a means for determining whether or not silt has been rapidly and recently deposited in an area, and the origins of the silt.

The coring tool was specially made for the purpose involved. Some of the cores require laboratory study and the coring tool was designed to permit removal and storage of a core in a few minutes time. The design of the corer finally developed is shown in the diagram, Fig. 2. The glass tube is held in place on a long handle by means of tubing clamps which can be released in about one minute by means of a screwdriver. At the upper end of the glass tube is a rubber stopper placed so as to form a valve. When the tube is forced into the mud, water is released through this valve. When the tube is withdrawn from the mud, the valve seats and retains the core plus the column of water over it. Very thin muds can be picked out of the bottom if a cork is inserted into the lower end of the tube before it is raised clear of the surface of the water. Moderate to stiff muds will remain in the tube without a stopper long enough to be brought aboard. Special racks were built for storage of cores in transportation. Cores were kept covered with water until removal from the tubes. Removal was accomplished by means of a pressure bulb or motor driven air pressure. Since the tubes were transparent, preliminary study was made by first washing the adhering mud from the outside of the tube, then turning a strong light on it. Fine structures, down to fine sand size, were located, and mapped on core diagrams before removal of the cores. After removal, cores were studied microscopically, in part or in whole, by dividing into sections (usually one

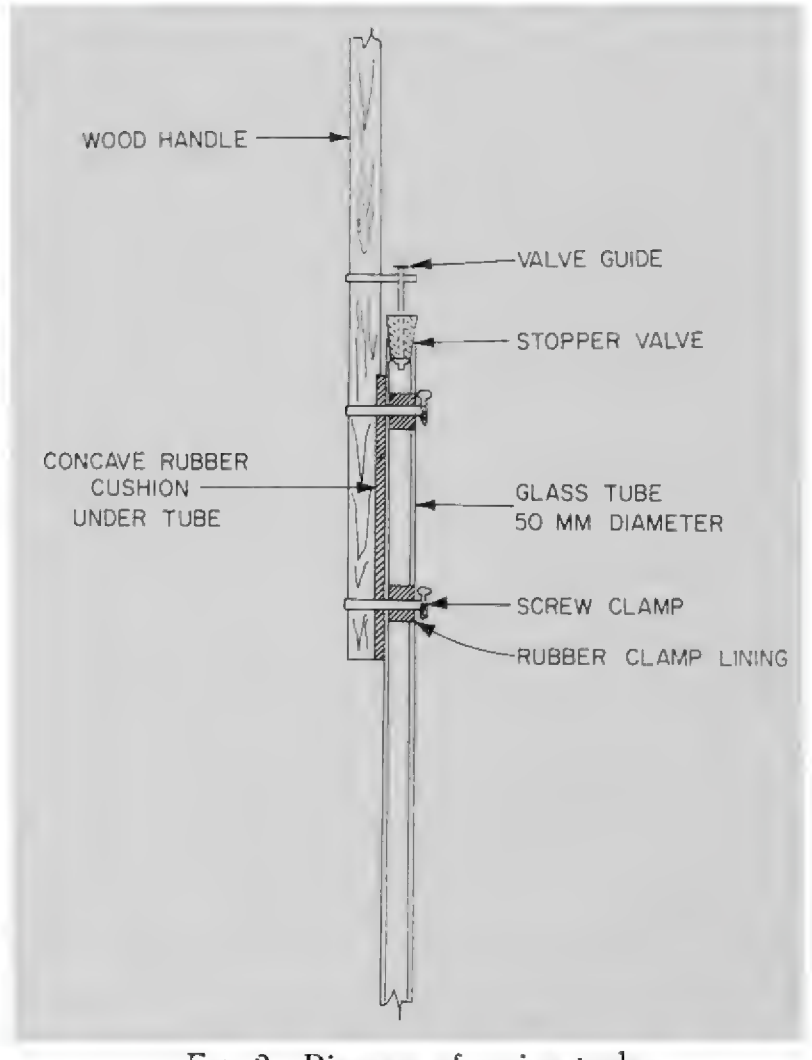

Fig. 2. Diagram of coring tools.

inch long). These sections were broken up in a large beaker of clear tap water and then poured through a nest of standard geologic sieves. All fine materials less than 1/200 inch in diameter are washed through the sieves and discarded. All particles of size larger than 1/200 inch are retained and sorted as to size by the sieves. The nest used usually contained sieves of 1/200, 1/150, 1/100, 1/80, 1/50, 1/30, 1/20, and 1/10 inch mesh.

Description and Hydrography of the Study Area

The Mississippi Delta is made up of mud deposits laid down by the river in a flat deltaic plain. The Delta is distinguished by the presence of distributaries, branches of the river in which the water flow is away from the river rather than toward it as in tributaries. The elementary steps in delta building are summarized by O'Neil (1949). The distributaries, overflowing their banks periodically, deposit the muds along their courses on either side and thus form natural levees. The marshes and their numerous ponds and lakes form between these natural levees. At the present time, the active portions of the Mississippi Delta are confined to the end of the river from the Baptiste Collette Subdelta southward on the east bank, and from Venice southward on the west bank. Otherwise the Atchafalaya is the only remaining active distributary; it is now building a delta. The Atchafalaya is the westernmost of the major distributaries. Its recent growth probably is due partly to pressure created by the Mississippi River levees and by the severing of the other major distributaries. The Atchafalaya is also a shorter route to the sea for Mississippi River waters.

Levees of the Mississippi River

The man-made levees along the Mississippi have altered the Delta area (Gunter, 1952). The area between the Mississippi and Bayou Lafourche now receives no silt from the Mississippi River during flood periods. The last significant addition of mud from the Mississippi occurred with levee breaks in 1927 (the Junior Crevasse).

The most far-reaching change produced by the levees has been to increase the salinity of the bays and bayous in the inter-distributary basins. Deprived of the fresh water from the Mississippi River, these water bodies often have excessively high salinities, approaching oceanic concentrations. The loss of the annual flushing action which formerly took place each spring during the river flood period is also a significant change. Diseases of oysters and the principal predators were annually partly eliminated from the bays by fresh water prior to the building of the levees (Mackin and Hopkins, 1962). Now, the fresh water is carried through the still active passes at the mouth of the river and dissipated in the Gulf. Some of this water returns to bays in the area from the Southwest Pass to Barataria Bay because of the westerly drift in the Gulf of Mexico in the inshore area west of the Mississippi. Loss of fresh water from the river is paralleled by loss of the sedimentary materials that normally overflowed into the bays. This effect will be discussed more fully below.

Organic Matter in Marsh Muds and Bay Bottoms

Bays developing in former marshlands have a high percentage of organic matter in the muds and bottoms. The source of most of the carbonaceous matter is the marsh,

which is built of deep layers of peaty materials, derived from stems and roots of marsh plants. The highest percentage of organic detritus occurs in the bay margins and in the areas where islands have been partially or wholly destroyed.

Deposition of mud and plant debris from eroding bay margins and islands, together with plankton production in the water, may counteract deepening. A bottom may gradually accumulate sediment from these sources, the older layers becoming more deeply buried in the bottom. The actual depth of the water therefore may not increase as rapidly as the rate of subsidence and rising Gulf level would indicate.

Tides in Delta Bays

Tides on the Louisiana coast are of the daily type with one high and one low level per day. The tides respond more closely to the declination of the sun and moon than they do to the moon's phases. Irregularities in the daily periods of low water and high high water result. The range of the tide varies (1) with the approximately bi-weekly period, (2) seasonally, and (3) over a long period of years.

In the Louisiana area the normal maximum range is about 1.2 to 1.8 feet at periods of tropic tides (at which time the moon's declination is greatest), and varies only 3 or 4 inches between high and low at equatorial tide periods. Tidal current velocity varies with the vertical height and differs from day to day in the bi-weekly cycle.

Mean low tide (and high tide) approximates 0.9 foot (nearly 11 inches) lower in January than in August. This makes navigation more difficult in winter.

Current Velocities in Bays, Bayous, and Passes

A number of measurements of velocity of water movement have been made. These are important since the distance to which silt may be carried is largely a function of current velocity. In most bays, water movement is slow, the mean rate rarely exceeds 0.5 knot, and the usual rate is of the order of 0.2 to 0.3 knot. Wind driven currents are usually of the same order of magnitude, except under conditions of storm or hurricane. In Table 1 are presented some representative measurements in parts of Barataria Bay and in Terrebonne Bay.

In some bayous movement is faster than in bays, with maxima often between one and two knots (Table 2). Such movement may produce scouring action, and bayous with high rates of water movement are generally deep compared with the bays. It is common to find bayous 10 to 20 feet deep connecting shallow silted bays whose depths do not

Table 1

Water movement in bays

Bay	Author	Rate of movement (knots)	
		Mean	Maximum
West side of Barataria	Austin, 1955	0.27	0.54
Upper Barataria—off end of Bayou St. Denis	Marmer, 1948	0.49	1.30
Upper Barataria Bay, 1.1 mile NE of Manilla Village	Marmer, 1948	0.45	0.90
Barataria, 2 miles SE of Manilla Village	Marmer, 1948	0.26	0.80
Terrebonne Bay	Marmer, 1935	0.35	0.50

TABLE 2

Current velocities and volumes of water movement in several bayous in Louisiana

Name and location of bayou	Author and dates	Water volumes, cubic feet per 24 hr period (millions)		Size of bayou		Current velocities, knots	
		Maximum flow recorded	Minimum flow recorded	Maximum depth feet	Width feet	Maximum	Mean
Bayou Lafourche, N of Leeville bridge	Lynch, 1949	80.4	58.5	10.5	220	1.0	0.4
Bayou Lafourche, S of Leeville Bridge	Lynch, 1949	128.4	57.0	11.5	206	1.2	0.5
SW canal E of Leeville	Lynch, 1949	94.5	39.3	9.5	120	1.6	0.7
SW canal W of Leeville	Lynch, 1949	43.8	21.6	8.5	115	1.0	0.4
Bayou Auguste at N end	Lynch, 1951	63.4	.....	.....	500*	0.9	0.5
Bayou Fontanelle, location 1	Lynch, 1951	36.9	.....	.....	150*	0.9	0.5
Bayou Fontanelle, location 2	Lynch, 1951	68.6	.....	.....	200*	.....	0.5
Dupre Cut	Lynch, 1948c	39.0	37.0	11.0	220	0.5	0.5
Manilla Bayou, station 1	Jung, 1957	425.1	123.3	18.0	220	1.4	0.9
Manilla Bayou, station 2	Jung, 1957	182.2	2.8	20.0	210	0.6	0.4
Rattlesnake Bayou	Lynch, 1948a	492.5	259.2	16.0	300	1.6	0.9
East Pass from Billet Bay	Lynch, 1948a	95.3	23.6	4.5	210	1.5	0.6
South Pass from Billet Bay	Lynch, 1948a	31.4	4.1	5.5	263	0.5	0.2

* Approximations.

exceed 3 or 4 feet. The current rate may be judged roughly by the depth of a bayou, as compared with bays in the same vicinity. Deep bayous are often bottomed with stiff clay, with silt only in the margins and the inside of curves. Such clay is generally only thinly covered with marine deposits, if these are present at all. Water borne deposits of silt do not occur in these scoured bayous.

On the other hand, there are many "dead" bayous. These are generally not more than 2 to 3 feet deep, and are invariably bottomed in deep, soft silt. Most of them are abandoned waterways, that is, abandoned by the water itself as a major route for tidal waters. Many of these are segments of formerly continuous bayous or distributaries, parts which became "drowned" in developing bays. An example is shown in the map (Figs. 1 and 6). This one originally followed a route through marsh across what is now the western end of Lake Grande Ecaille. As Grande Ecaille expanded it broke through the natural levees of this bayou, leaving both upper and lower ends dangling and without current. Dead bayous have only enough current to fill and drain them by tidal rise and fall, and there is little or no through movement.

Major water movement takes place through passes to the Gulf, with consequent higher maximum velocities of water flow (Table 3). Barataria Pass has a maximum rate of nearly three knots and the mean flow is well over one knot. Barataria Pass is scoured to a depth of more than 165 feet in one place and carries a very large volume of water in and out. Other passes do not approach Barataria in depth, but Cat Island Pass (into Terrebonne Bay) carries much more water because of its great width. Small passes may become sand-choked, as for example, Pass Abel.

NATURAL TURBITIES AND SEDIMENTATION

Hewatt (1949) made extensive measurements of turbidities in Louisiana bays, especially Barataria and its satellite bays. He measured turbidity by means of photo-

TABLE 3

Current velocities and water volumes in several major passes in Louisiana
Data from Marmer, 1935, 1948

Pass	Rate of movement		Volumes per daily tide cycle (in feet)
	Mean	Maximum	
Barataria position 1	1.3	2.9	2887×10^6
Barataria position 2	1.2	2.8	.....
Barataria position 3	1.2	1.6	.....
Quatre Bayou	1.1	2.2	1879×10^6
Pass Abel	1.2	2.5	341×10^6
Caminada	1.5	2.4	1280×10^6
Wine Island	1.3	2.3	.....
Cat Island	0.6	....	15117×10^6

electric cells which recorded light transmission through a column of water pumped through a pipe. These machines were mounted in stationary stations at various points in Barataria Bay and in surrounding adjacent bays and bayous, or were mounted in boats. In the latter the intake pipe was set just forward of a skid which held it about one foot over the bottom.

Hewatt's maximum turbidity measurement was nearly 900 ppm (see summary by Mackin and Hopkins, 1962) in mid-lower Barataria Bay. Such high turbidities were not of long duration, a matter of hours producing drastic decreases (and increases), and are probably not significant beyond designation of maximum values. Minimum values were between 5 and 10 ppm.

In Fig. 3 are presented turbidity frequencies for four months at a lower Barataria

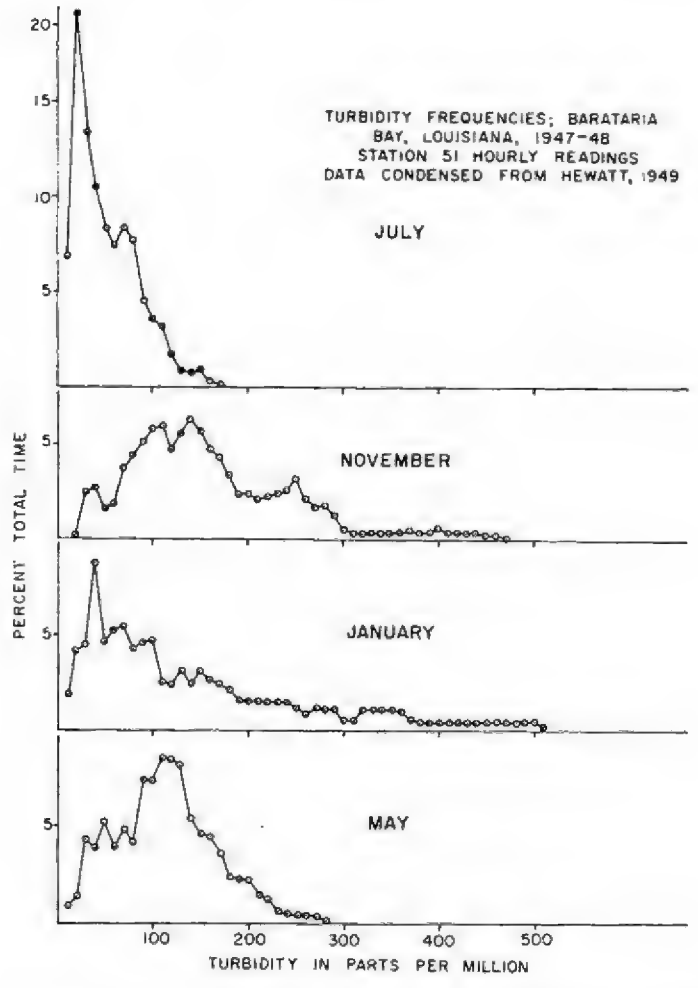

FIG. 3. Turbidity frequencies; Barataria Bay, Louisiana, 1947–48, Station 51, data from Hewatt (1949).

Bay station, condensed from Hewatt's data. In July, most of the values were in the range of 20 to 60 ppm; less than 5 percent of the values exceeded 100 ppm and none attained 200 ppm. In November, values between 100 and 200 ppm predominated; those between 200 and 300 ppm were next in order; and turbidities measuring up to 450 ppm were common. The January record showed even wider distribution with a range from 10 to more than 500 ppm. In May there was a tendency toward return to the low readings of summer, but the mode was still above 100 ppm.

Seasonal variations in turbidity are shown in Fig. 3, with summer minima and winter maxima, the latter associated with a higher percentage of northerly winds and cold fronts. Water level is lowest in January and highest in late August and September (Marmer, 1954). It is proposed that turbulence created by wind is effective in stirring up bottom muds in inverse relation to depth especially where waves from deeper water shoal. A lower mean tide level means a significant reduction of water depths, and north winds reduce tide levels still farther at the time of lowest mean tide level. It follows that shallow bays, and shallow marginal areas of large bays, may be more turbid than deeper ones even though a shallow bay may also be smaller with less wind fetch.

Observations in the Louisiana area show that high salinity areas to the south are less turbid than the low salinity areas to the north. The electrolytes in sea water flocculate clay and fine silt. Also the high salinity areas are also usually the deeper, more southerly parts of bays with more coarse sand and less fine soft materials. There is more admixture of clear Gulf water from the south also.

Local turbidities may vary sharply within a very short distance. This is illustrated in Fig. 4, which is a record of 28 turbidity measurements made while drifting a distance of roughly 2700 feet. The boat passed successively over (1) a shallow silty flat, (2) a shell reef, (3) a shallow inshore area, and (4) a deeper chanel through which flowed

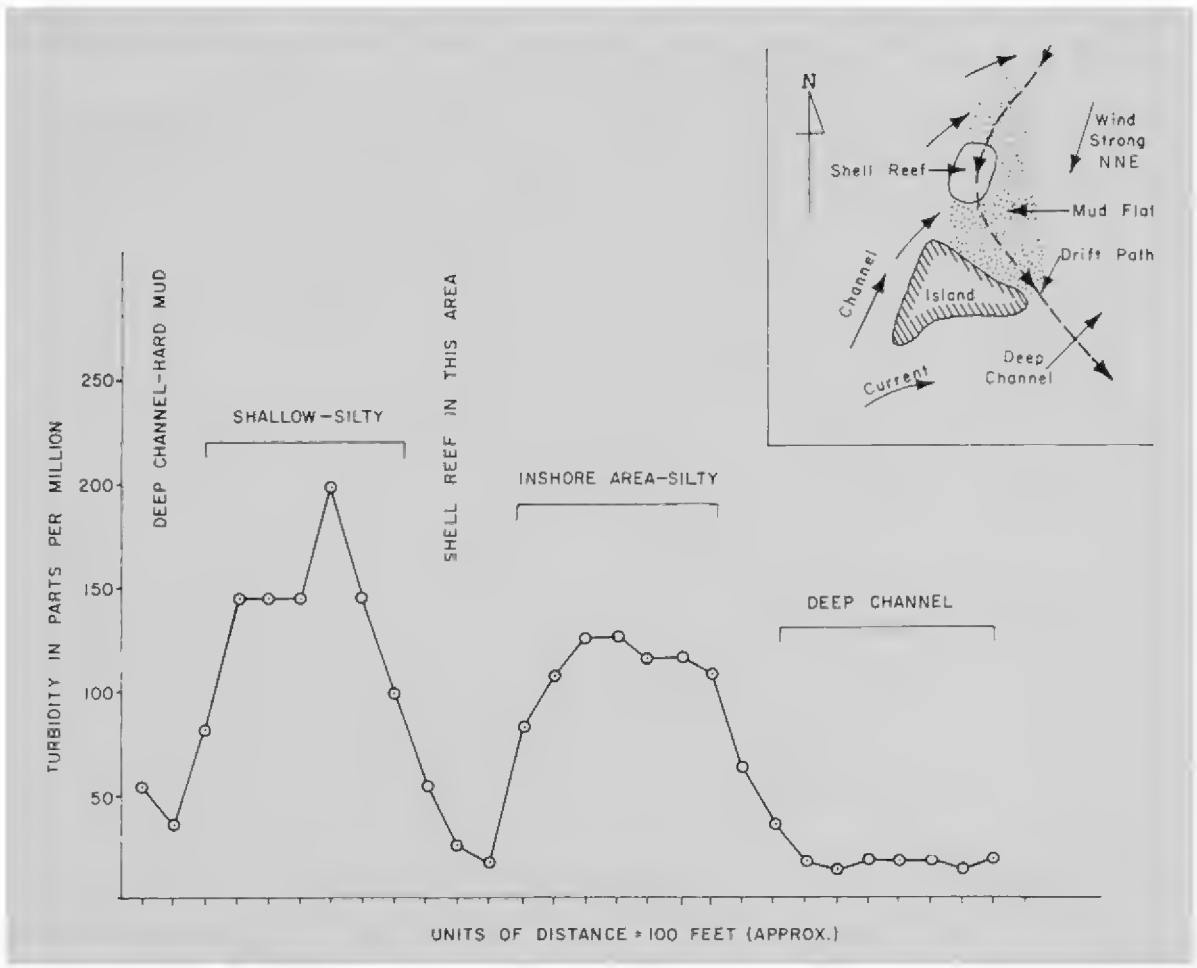

FIG. 4. Effect of type of bottom and depth on turbidity.

high salinity water on flood tide. Turbidities varied from a maximum of nearly 200 ppm to a minimum of about 12 ppm. Such radical variations are unusual, in such a small area, but variations of less magnitude are quite common.

Subsidence of the Delta and Rising Sea Level

All of the northern Gulf shore is said to be sinking and in the Delta area the effect is enhanced by compaction of the muds. The magnitude of the subsidence varies in different parts of the marsh. Gunter (1952) gave estimates from varying sources which indicate that the rate of settling ranges from 0.05 foot to 0.163 foot per year in the Delta area. The greatest sinking rate is probably at the end of the active Delta, where compaction plays a greater role than elsewhere. If we assume a mean of 0.1 foot per year, in a 10-year period the bottom of a bay would drop out by a foot. The sea level is also rising (Marmer, 1954). This author gave a figure of 0.03 foot per year at Pensacola, Florida, but it is not known what part of this was actual sea level rise and what part was land subsidence. If the sea level rise is additive to subsidence, Louisiana bay bottoms must be accumulating sediments at a comparatively rapid geological rate in order to maintain the status quo so far as water depth is concerned. However, there is strong evidence that sedimentation in most areas does not keep up with subsidence and that the bays are actually deepening.

Studies of foraminifera in the bottom muds often indicate concentrations far beyond what would be possible if siltation is taking place at a rate of 0.1 foot per year. Actual depth measurements also show an increasing depth in parts of Barataria Bay and Lake Grande Ecaille (Lynch, 1948b). In Fig. 5 are comparisons of depth in Grande Ecaille for 1934, 1948, and 1959. The 1934 data are from soundings by the U. S. Coast and Geodetic Survey. In 1948, Lynch made soundings over the 1934 data lines, and in 1959 these same transects were sounded again. All depths were reduced to mean low tide levels. Increased depths of up to 5 feet are indicated in the channel areas and a little more than one foot in most of the bay. The transects (see Fig. 6) pass nearly over the locations where foraminifera in the thousands were found in the muds in February, 1956. These abnormally large numbers may indicate that concentration is taking place by a winnowing process.

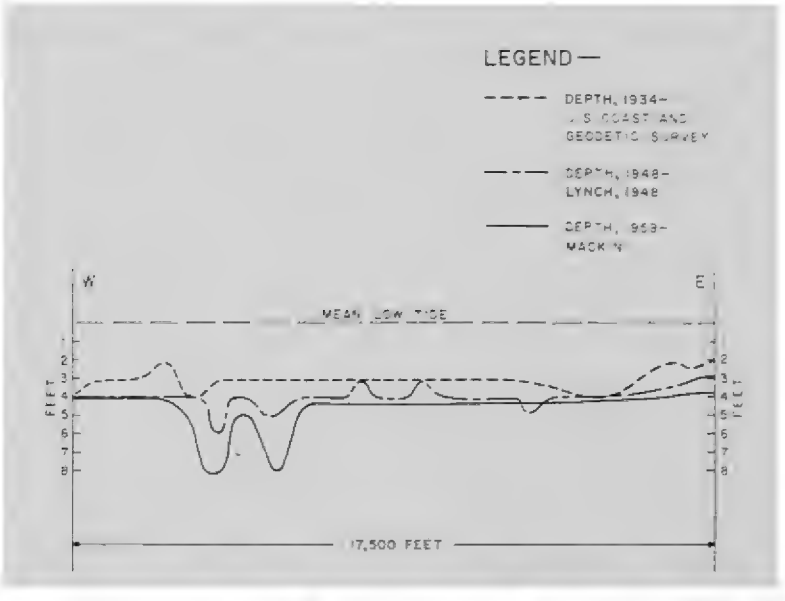

Fig. 5. Depth transects of Lake Grande Ecaile, showing depths in 1934, 1948, and 1959. Data for the transects of 1934 and 1948 are from Lynch (1948a).

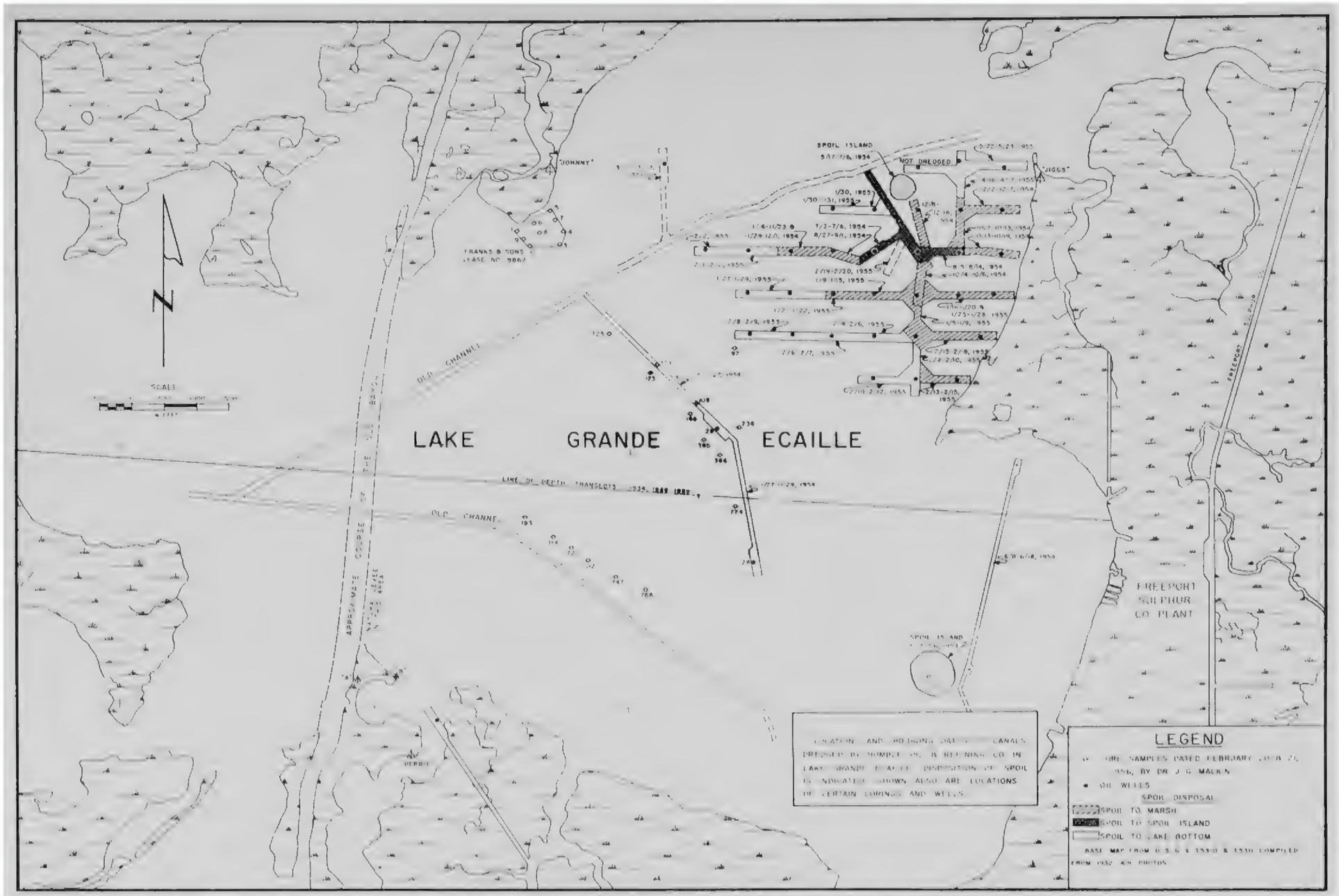

FIG. 6. Map of Lake Grande Ecaille, showing canals dredged by Humble Oil and Refining Company, and indicating the disposal method for spoil and dates of dredging.

O'Neil (1949) states that subsidence in "inside" ponds, in fresh water, and in nearly land-locked situations takes place at a rate of 4 to 6 inches annually where no silt accumulation occurs. Owen (1955) listed a number of evidences of subsidence, and discussed at length the changes produced in the marsh land by the levees of the Mississippi especially the elimination of new silt. Gunter (1952) mentioned the diversion of silt and Lynch (1948b) assumed that no new sediments were now being added in the Barataria Basin, but that existing silt was being reworked. This interpretation is probably approximately correct, even though it fails to consider the biological production of organic substances *in situ*, which now make up more and more of the new sediments. The increasing peat deposits indicate that organic materials may not decay as rapidly as they are formed. The data do indicate that addition of sedimentary materials to those existing prior to the completion of the levees is now small, if not negligible.

Disintegration of Shorelines

As Lynch (1948b) showed, depths of bays may be influenced by relocation of existing sediments. When shorelines disintegrate, the materials are deposited in the bays. Wasting away of marsh areas, islands, and shorelines has been one of the outstanding phenomena of the past 60 to 70 years. Owen (1955) stated that the water to land ratio increased by 25 percent in that period due to rising sea level and marsh disintegration. Complete obliteration of some small islands has been observed in the last 10 years, along with extensive caving of shores, and sinking of marsh areas to form new ponds.

Much of the marsh is now in an unstable semi-suspended condition floating on a semi-fluid muck, and the marsh surface is barely inches above sea level due to subsidence of the clay-pan under the marsh. Healthy growth of marsh plants is often prevented by abnormally long periods of submersion in highly saline water in summer. When marsh plants die their roots cease to bind the marsh silts, accelerating the break-up. Exposed shorelines are undercut by wave action and the marsh plants may sink into the water in large blocks.

Transport of Turbid Materials from Bays to the Gulf

Since a very great volume of turbid water in Louisiana bays leaves the passes with each ebb tide, a certain amount of suspended material also passes outward. About 4.3 billion cubic feet of water goes out through Barataria Pass each tidal day. A considerable part of the silt load of this water is dropped in the Gulf because (1) the current decreases as the water reaches the Gulf and (2) the higher Gulf salinity produces a greater flocculation effect. Excess of ebb over flood is about 200 million cubic feet of water. It is estimated that this water, which does not return to the bay on flood tide, removes about 620 tons of silt on a day when the mean turbidity in the bay is 100 ppm. This is only a small part of the daily removal, counting that dropped by the 4.3 billion cubic feet. If 10 percent of the load is dropped and the turbidity is 100 ppm the amount would be 1342 tons. However, a similar amount is brought into the bay from inflowing tributary bayous, and the amount lost would merely be a rough estimate of the amount of silt moved through the bay under the conditions named. The figures given are to indicate order of magnitude only. No claim of accuracy is made. It is

clear that the transport of suspended materials may be measured in thousands of tons daily.

Volumes of Turbid Materials Moving Over Oyster Beds Under Natural Conditions

In evaluating the effect of dredging operations in silting of oyster leases, it is instructive to determine what are the volumes of silt in suspension which normally pass over the beds in a unit period. When the mean turbidity is 100 ppm, the mean current velocity 0.3 knot, and the open bay 5 feet deep, the amount of silt passing over the bed per day can be roughly calculated per 200 ft width of bed at right angles to the current direction. The calculation follows with conversions: 6080 ft per nautical mile; 24 hr per day, 62 lb of water per cubic ft.

$$\frac{(0.3 \times 24 \times 5 \times 6080 \times 62 \times 200)\ 100}{1{,}000{,}000}$$

$$= 271{,}400 \text{ lbs. or } 135.7 \text{ tons } (123{,}105) \text{ kilograms}$$

When considering the effect of suspended silt on an oyster bed, it is well to inquire as to the reason why such weights of natural silt do not fill all the bays in a short time. The answer is that the silt has its origin, for the most part, in the bay bottom itself. It is merely picked up and put in suspension by turbulence, carried laterally for a greater or less distance and dumped back on the bottom. It is a part of the progression of steps by which silt ultimately ends in the sea.

Depth Equilibrium in Bays

Bottoms of deltaic bays are generally comparatively soft, especially in the Mississippi Delta where levees prevent deposit of coarser materials in most bays at the present time. Sand encroaches through the passes from the Gulf (Rasmussen and Lynch, 1949), but most of the presently accumulating sediment originated in the marshes of the Delta itself. To some extent the sediments forming the present bottoms of the bays such as Barataria are merely resorted deltaic deposits with predominance of finer particle sizes. A very high proportion of present bottom materials is peat, derived from the marshes themselves. In some cores more than 50 percent of the material is ignitable. Crumbling of shorelines and deepening and widening of passes tend to increase water areas at the expense of the marshes. So far as depth and size are concerned the bays approach an equilibrium. Over short periods of time (weeks, months, or a few years) any given area may be in equilibrium between deposit and scour, irrespective of the amount of silt which may be transported over a section of bottom by water. The amount of material suspended in the water has little to do with scour or fill. The depth of any area is primarily controlled by the rate of movement and volume of the water. Any change in current rate will produce an immediate scour or fill.

There are exceptions, such as oyster reefs, where increase or decrease in current may produce no effect at all. Such biologically controlled bottom materials are not moved by current, since the shell mass is too heavy, but may be covered with soft silt if a decrease in current occurs. Where shell is found in the surface muds of the bottom but not in a continuous reef, changes in depth may occur as if the shell were not there, by

winnowing of the fine sediments as currents increase, or by sedimentation as currents decrease. Winnowing concentrates shell materials in the surface layer of the bottom.

Weights of Suspended Matter in Water

One cubic foot of water weighs about 62.43 lbs. One cubic foot of sea water weighs more, at salinity 20 ppt, about 1.25 lb more with the total weight per cubic ft about 63.68 lb. However, with the procedure used for estimating turbidity it is assumed that the bay water weighs the same as distilled water as if the water had no salt.

If the turbidity is 10 ppm, the weight of suspended matter in one cubic foot of water is 0.0006243 lb (0.0000283 kg). For expansion of these relations see Table 4. In a square (nautical) mile of water, turbidity 100 ppm, 5 feet deep, the total weight of suspended matter is:

1,153,906 lb = 576.95 short tons (about 523 metric tons)

There is considerable error if the turbidities as colibrated against fuller's earth are used for calculating weights of suspended matter in Louisiana bays. Our studies have shown that in Louisiana bays about 0.7 as much silt (by weight) will produce a turbidity reading equivalent to 1.0 weight of fuller's earth (the turbidity standard material). The specific gravity of the Louisiana silts is less due to the high organic content. It is necessary, therefore, to apply a factor of 0.7 to all results, and the figures given above for the weight of suspended matter per square mile become:

807,734 lbs. or 403.86 tons (about 366 metric tons).

The volume of this weight of suspended matter is

$$\frac{(807{,}734/62.43)}{1.5} = 8625.5 \text{ cubic feet (about 244 cubic meters)}$$

Settling Velocities of Particles of Various Sizes

The distance to which particles of various sizes may be carried by currents depends partly on the settling rate through water. Stokes developed an equation for determining settling velocity of spheres. Wadel and Stokes, using a modified equation, calculated the settling velocities of quartz spheres in the size range from very coarse sand to clay

Table 4

Weights of suspended materials in one cubic foot of water at various turbidities

Turbidity, ppm	Weight of silt in one cubic foot of water (pounds)
10	0.0006243*
20	0.0012486
30	0.0018729
40	0.0024972
50	0.0031215
60	0.0037458
70	0.0043701
80	0.0049944
90	0.0056187
100	0.0062430
200	0.0124860
300	0.0187290

* 0.0000283 kg.

particles of very small size as given in Table 5, modified from "The Oceans" by Sverdrup, Johnson and Fleming (1942).

Particles of shell larger than 1 mm fall through 5 feet of distilled water in a few seconds. Those as much as 1/10 inch in diameter drop through water so rapidly that a strong current cannot carry them more than a foot or so before they hit bottom. Most foraminifera sink at a rate between that of coarse and medium sand. All sizes fall through saline water slightly more slowly than through distilled water, and turbulence may keep particles in suspension which would otherwise drop to the bottom. According to the table the finest clay particle may be carried for more than 6000 miles before it sinks one foot in a one knot current. However, flocculation in saline water may precipitate such clays in a relatively short period.

Particles on the bottom may be rolled provided the current rate is sufficient and the bottom water movement is turbulent. Current rates through some passes are sufficient to roll or carry sand from the Gulf of Mexico to positions a considerable distance inside the bays. However, most sand particles found in the bays were brought in during river overflow periods prior to completion of the levee system. Sand is still deposited near the several presently active Mississippi sub-deltas.

Effects of Dredging on Turbidity

Several studies were made on the drift of spoil away from working dredges and the results will be presented. Other things being equal, the distance to which spoil may be carried away from a dredge is a function of the current rate. Some particles are heavy enough and large enough that they never move from a spoil area. However, some clays and silts from a dredge become part of the general turbidity of the water, and the materials become a part of the multi-stepped movement to the sea. Only when the concentrations of silt become much greater than normal are the materials directly deposited on the bottom as spoil.

The next section deals with the volumes deposited at different distances from a dredge.

Turbidities and Turbid Drift from a Working Hydraulic Dredge in Eastern Lake Grande Ecaille

In the period from June 8 to June 16, 1954, a hydraulic dredge cut a canal from a point in southeast Grande Ecaille in a northerly direction to a well site about 4500 feet away. For the location of this canal see the map, Fig. 6.

Table 5

Settling velocity of quartz spheres in distilled water at 20°C

Size of particles	Diameter mm	Time to fall 10 cm.			
		Days	Hours	Minutes	Seconds
Very coarse sand	1	....	....	.. .	0.6
Medium sand	1/4	....	....	....	2.7
Very fine sand	1/6	....	....	.. .	29.0
Silt	1/32	....	.	1	55.0
Silt	1/256	....	2	2	32.0
Clay	1/512	....	8	10	.....
Clay	1/8192	87	3	19	.....

All spoils from this operation were deposited in a "spoil island." The end of the discharge pipe was fixed in position, about eight feet over the water. About two days were required before the build-up of spoil under the end of the discharge pipe reached the water surface. During these two days turbidities produced at the discharge site were very high and the distance of travel away from the site was relatively great. After the apex of the spoil island emerged from the water, turbidities were radically reduced as the spoil was then discharged on "land" and rolled or was washed into the surrounding water.

At each station samples for turbidity checks were taken with a Foerst sampling tube at the surface and approximately one foot over the bottom. Studies were made daily until the dredging operation was completed.

Locations for sampling were determined by the drift of current away from the spoil outlet. Direction of this drift was determined on each sampling date. The samples were taken at intervals from the dredge to the greatest distance that turbidities were found higher in value than the daily controls.

Control samples were taken on each day of the study. Some of these were taken in Barataria Bay, in Cat Bay, and in western Lake Grande Ecaille, 2 to 10 miles from the dredging operation. Others were taken in the vicinity of the spoil outlet, but up-current from the outlet at a distance sufficient to be certain that no turbid materials from the dredging were taken. These latter controls were most useful because depth, wind fetch, and bottom character were similar.

The data derived from this study are presented in Table 6 and Fig. 7. The greatest distance to which spoil was transported was about 1300 feet. On most days (after the spoil island was emergent) turbidities were negligible at 100 feet from the discharge pipe. Spoil rolled down the slope of the spoil island but failed to increase turbidity measurably at one foot over the bottom. Tidal currents in this area (southeast Lake Grande Ecaille) were too weak to be detectable. It was noteworthy that spoil drifted away in directions from east around counter-clockwise to the northwest, but there was no drift in a southerly or southwesterly direction.

Maximum turbidity measured in this study was 1980 ppm at about 30 feet from the end of the discharge pipe and down-current from the discharge. The highest reading at 100 feet was 1060 ppm. There were no readings as high as 100 ppm at distances greater than 300 feet. No readings greater than those observed at one time or another under natural conditions were observed at distances greater than 100 feet, but there are no data on turbidity between 100 and 200 feet from the spoil outlet.

A considerable number of control samples were taken during this study. The values of these ranged from 16 ppm to 97 ppm. These control samples were mostly from stations in Cat Bay and Barataria Bay. There were several of these which were not "natural" turbidities but were taken in the vicinity of shrimp trawlers. On a day when six "natural" turbidity samples ranged in value from 17 to 29 ppm, the turbidity in the vicinity of three shrimp trawlers was 71 to 97 ppm. On another date most of Cat Bay was made somewhat more turbid by trawlers than was Bay Ronquille on the east, or Barataria Bay on the west. In the vicinity of one trawler a reading of 75 ppm (surface) and 92 ppm (bottom) was made. This compares with a maximum of 39 ppm in Barataria Bay and 36 ppm in Bay Ronquille on the same date. The turbidities produced by the shrimp trawlers were not excessive, but they were greater than those produced by the dredge at distances of 300 feet from the spoil outlet.

TABLE 6

Turbidities around the spoil pipe outlet in ppm. East Lake Grande Ecaille canal
June 8 to June 16, 1954

Date	Turbidity; down current	Distance from spoil outlet; feet†	Turbidity; up current	Distance, feet†	To left of spoil outlet	Distance, feet†	To right of spoil outlet	Distance, feet†	Average turbidity; distant controls
6–8	864/1980‡	35	40/38	30	30/38	75	—/70	75	.....
	56/82	300	.....	.....	.....	.....	.....	.....	.....
	—/82	600	.....	.....	.....	.....	.....	.....	.....
	22/38	750	.....	.....	.....	.....	.....	.....	.....
6–9	660/1060	100	46/34	100	76/131	100	45/116	100	33/34
	45/59	500	.....	.....	.....	.....	35/34	400	.....
	56/45	1000	.....	.....	.....	.....	25/26	700	.....
	38/42	1300	.....	.....	.....	.....	17/20	900	.....
	28/30	2000	.....	.....	.....	.....	.....	.....	.....
6–10*	31/37	100	22/27	100	.....	.....	.....	.....	22/26
6–11	32/32	100	18/32	100	23/30	100	30/30	100	35/41
	35/36	600	22/22	1300	.....	.....	.....	.....	.....
	27/26	800	.....	.....	.....	.....	.....	.....	.....
	46/26	1200	.....	.....	.....	.....	.....	.....	.....
6–12	50/59	100	42/38	100	59/54	100	67/84	100	39/34
	50/59	600	.....	.....	.....	.....	.....	.....	.....
	56/50	800	.....	.....	.....	.....	.....	.....	.....
	56/59	2200	.....	.....	.....	.....	.....	.....	.....
6–13	94/108	100	45/42	100	61/67	100	48/46	100	41/40
	32/34	400	.....	.....	.....	.....	.....	.....	.....
	28/28	600	.....	.....	.....	.....	.....	.....	.....
	32/40	1000	.....	.....	.....	.....	.....	.....	.....
6–14	48/48	100	48/48	100	48/52	100	48/32	100	31/33
	28/28	400	31/31	300	.....	.....	.....	.....	.....
	27/31	600	.....	.....	.....	.....	.....	.....	.....
	28/28	800	.....	.....	.....	.....	.....	.....	.....
6–15	118/118	100	42/48	100	48/48	100	42/42	100	38/44
	42/42	200	.....	.....	.....	.....	.....	.....	.....
	25/26	400	.....	.....	.....	.....	.....	.....	.....
	25/25	600	.....	.....	.....	.....	.....	.....	.....
6–16	32/32	100	30/30	100	30/30	100	38/38	100	32/33
	24/24	300	.....	.....	.....	.....	.....	.....	.....
	25/25	500	.....	.....	.....	.....	.....	.....	.....

* About one hour after shut down.
† Distance from the spoil pipe.
‡ Surface turbidity left of slant; bottom turbidity to right.

Tidal currents in this part of Lake Grande Ecaille were negligible. The water movements were controlled almost exclusively by wind. Due to shifting wind, direction of movement of the spoil changed daily or more often. Preponderance of southeasterly winds produced drifts to the north and northwest on most days, but with the significant exception of three days of westerly winds (see Fig. 7). This westerly wind was very strong on June 12, and caused corresponding increase in distance to which suspended materials were transported toward the east.

The spoil island formed from this operation was about 1200 feet across. The apex was one to two feet above the high water mark and the center of the island was mostly composed of shell, which, being heavy, was not moved far from the spoil outlet. The margins of the spoil island were of fine soft silt. This material was mostly rolled down the slope of the spoil, and it is interesting that transport of significant quantities of spoil by this means exceeded transport of the suspended silt, both in distance and in amount.

Examination of this spoil island in 1956, two years after its formation, showed it to be relatively intact. Removal of materials by wave wash had leveled the apex, and

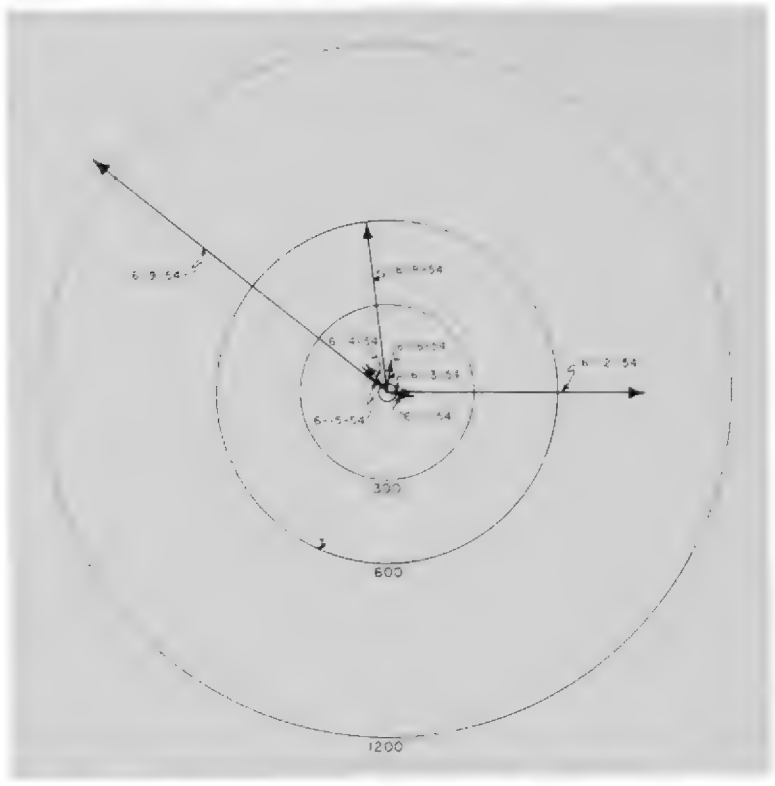
DIRECTION AND DISTANCE OF MOVEMENT OF SUSPENDED MATERIALS FROM THE OUTLET OF A SUCTION DREDGE IN OPEN BAY. SHORT ARROWS INDICATE THAT SPOIL IS BEING DEPOSITED ON THE APEX OF A SPOIL ISLAND OUT OF WATER, EXCEPT ON 6-11-54, WHEN THERE WAS NO DETECTABLE CURRENT. ARROWS INDICATE DIRECTION AND DISTANCE OF MOVEMENT OF SUSPENDED MATERIALS.
300
600
1200

FIG. 7. Direction and distance of movement of suspended materials from the outlet of a hydraulic dredge in eastern Lake Grande Ecaille.

compaction of the soft marginal muds had taken place. Otherwise, the amount of spoil contained seemed to be about the same as when deposited.

TURBIDITIES PRODUCED BY A CLAM-SHELL DREDGE

From January 8, 1956 to January 11, 1956, a clam-shell dredge was engaged in digging a slip for a well rig in the southwestern part of Lake Grande Ecaille. This dredge approached from the marsh (Fig. 6) and emerged into the bay proper at about 0730 on January 8. The southwestern margin of the slip was dug first, on January 8 and 9, and the northeast edge was then excavated, the dredge ending up in the early afternoon of January 11 at the northeast corner.

During all of this operation the wind was from a northerly direction and on the first day was strong. Tidal currents entered the cut (Fig. 8) directly west of the end of the slip at flood tide and moved eastward hugging the shore line. Most of the ebb tide periods were at night when the direction of movement was reversed but these movements were not observed. The movement of turbid materials from this dredge was followed to determine whether or not silt would affect nearby oyster leases. The corner of the nearest lease was about 200 feet to the northwest of the end of the slip. To the east, the nearest lease was about 1300 to 1400 feet away. This dredging offered an excellent opportunity to observe movement of water-borne materials away from a clam-shell dredge.

Three periods of observation were selected as representative of the conditions while the dredge was working. Minimum turbidity occurred while the dredge was just emerging from the marsh and maximum turbidities when the dredge was near the end of the slip, which occurred twice since the dredge cut the western half of the slip, then backed into position at the marsh edge again and cleared the east half of the slip. Fig. 8 shows

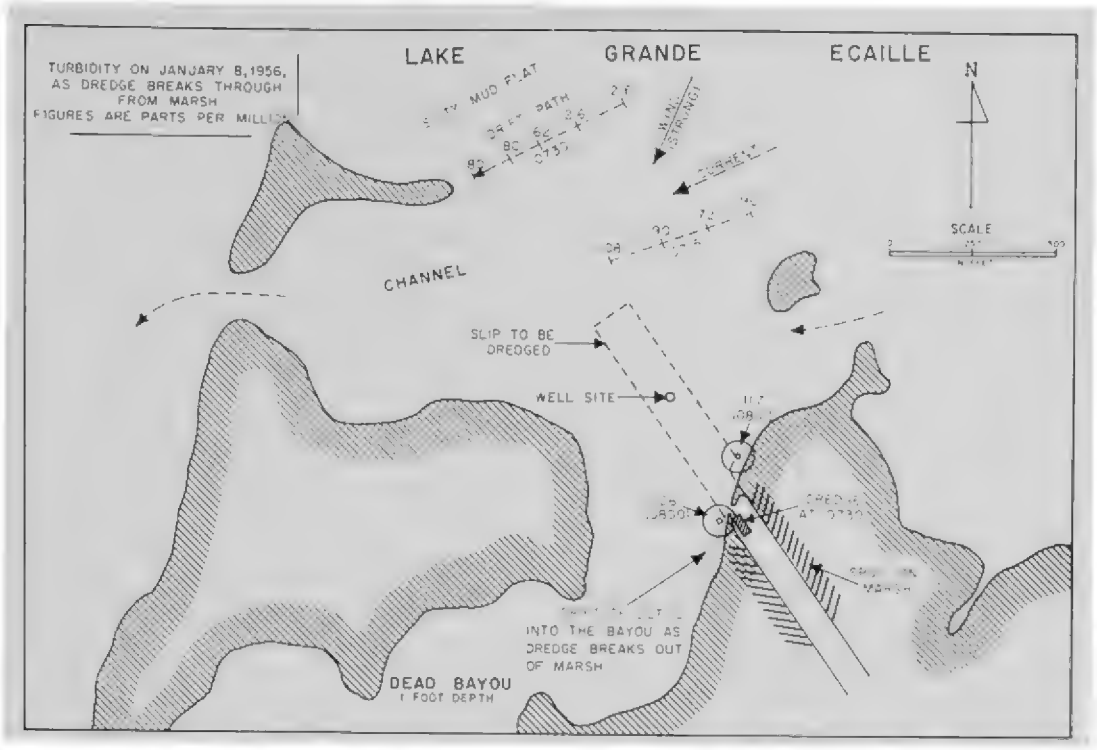

FIG. 8. Turbidity produced by a clam-shell dredge as it emerged from the marsh into Lake Grande Ecaille on Jan. 8, 1956.

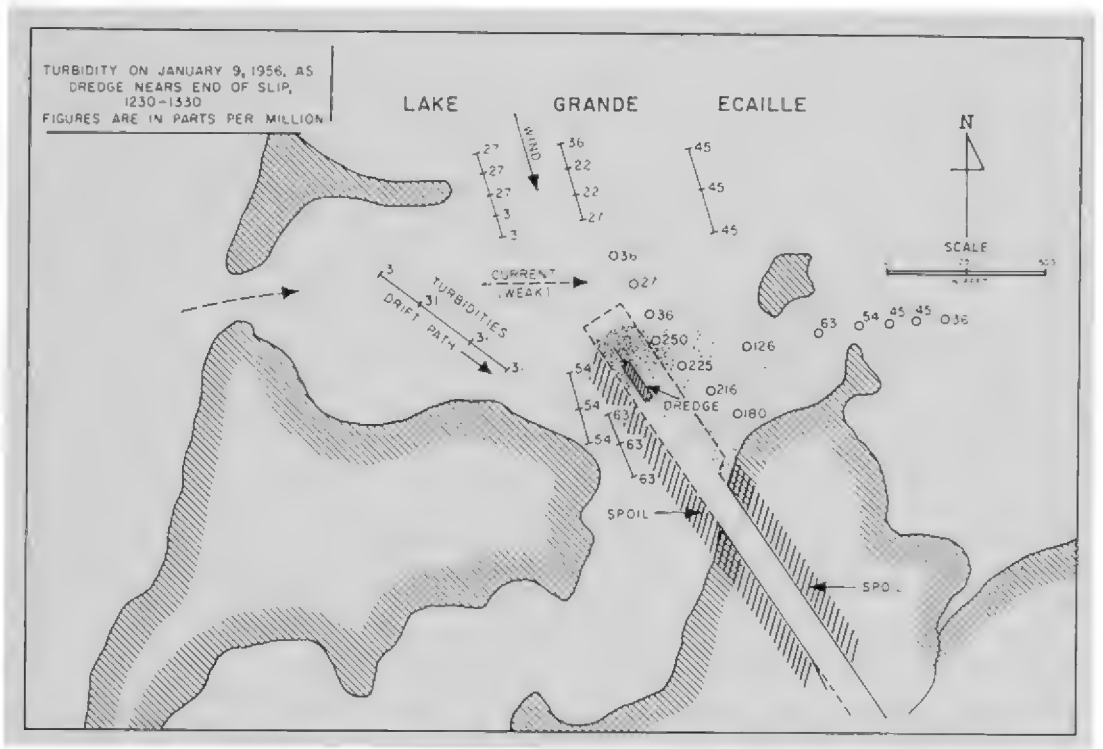

FIG. 9. Turbidity produced by a clam-shell dredge Jan. 9, 1956 as it completed the west side of the slip.

conditions at the time the dredge was emerging from the marsh. During the time that the dredge worked near the marsh edge, and partially in the shallow "dead" bayou, the wind drifted all the suspended silt into the bayou. During this period turbidity values were less than 200 ppm 100 feet from the dredge. Some natural turbidities observed were higher than those produced by the dredge.

When the dredge was near the outer part of the slip, tidal movement dominated the movement of suspended silt away from the dredge. January 8 to 11, 1956, coincided with a tropic tide period. Tidal amplitude was about 8 to 10 inches with low tide in the morning for the four days. During most of the day tide was flooding, moving suspended silt to the east, while the north wind kept it close inshore. Figs. 9 and 10 show the periods of maximum observed movement away from the dredge.

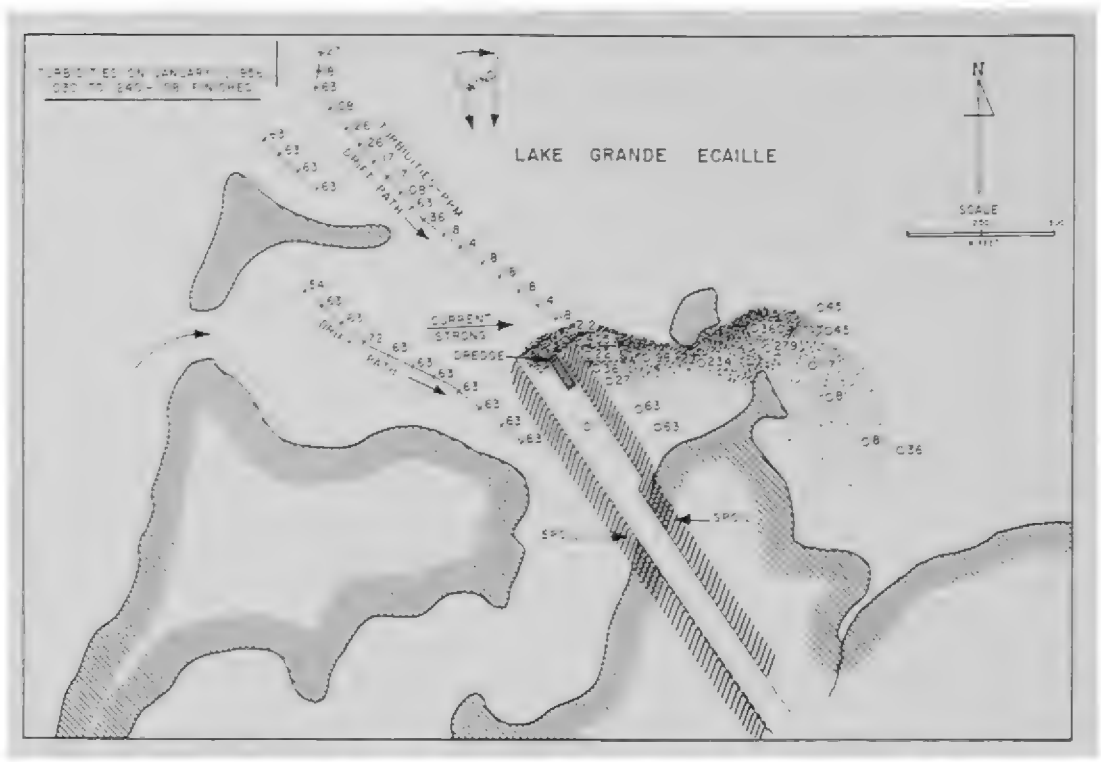

FIG. 10. Turbidity produced by the clam-shell dredge as it completed the east side of the slip.

Lines on Figs. 8, 9, and 10 represent approximate drift lines of the boat while taking samples, and the adjacent numbers are values of samples in ppm taken about one foot over the bottom. Closest observations to the dredge were at about 100 feet. The highest turbidity recorded was about 400 ppm about 100 feet to the east of the dredge on January 11. Stippling on the figures indicates the area estimated by sampling to be affected by the dredge. On January 11, some effect was still measurable about 1200 to 1300 feet away. However, note that just above the boomerang-shaped island, the wind was producing turbidities up to 126 ppm on the silty mud of lease 9864.

If we assume that the turbid area shown in Fig. 10 with 200 ppm or greater of suspended material received 200 ppm of silt during one-half of the dredging period, and that this much silt dropped out on the average each 30 minutes (the approximate time required for the current at 0.3 knot to move the materials across the 1000-foot long area), the depth of the silt, if all stayed where it landed on the bottom, would have been about 1/2 inch.

HYDRAULIC DREDGE IN NORTHWEST LAKE GRANDE ECAILLE

On March 22 and 23, 1956, the Humble Oil and Refining Company dredged a short canal and well slip in northwest Lake Grande Ecaille (map, Fig. 6). A hydraulic dredge was used and the spoil was spread on the bottom east of the canal right-of-way in a band about 350 feet wide, the nearest part of which was about 50 feet from the canal margin. The discharge pipe was mounted on a small barge. Discharge was into the air in a half-fan circle. The discharge barge was moved from time to time to prevent build-up of the bottom greater than six inches.

The Fisher Electrophotometer was used to map the turbid waters caused by the discharge. Direction of movement of the suspended silt was established by visual examination, by floats, and by sampling in a circle around the dredge discharge barge. The positions of the dredge and discharge barge and the relation of the canal to the buoyed channel, at the period of measurement on March 22, are shown in Fig. 11.

On that date, general turbidity in the Lake was fairly high, about 60 to 150 ppm,

depending on the location of control samples and the time of day the controls were taken. In the area upstream from the discharge, control turbidity values were about 80 to 150 ppm (Fig. 11).

Turbidities in the area within 100 feet downstream from the discharge were in the range from 1000 to close to 2000 ppm. Two samples at approximately 100 feet showed about 1500 ppm turbidity (Fig. 11).

The suspended material on March 22 was drifting away in a general northerly direction. Numerous samples were taken while drifting the boat through the turbid area and the general shape, length, and lateral spread of the turbid water was roughly mapped as shown in Fig. 11. The greatest distance that the suspended material drifted on that date was about 1200 feet; the lateral spread was nearly 700 feet.

On the following day, March 23, with the canal nearly finished, the drift was generally to the southwest and the greatest length of the turbid area was between 800 to 900 feet

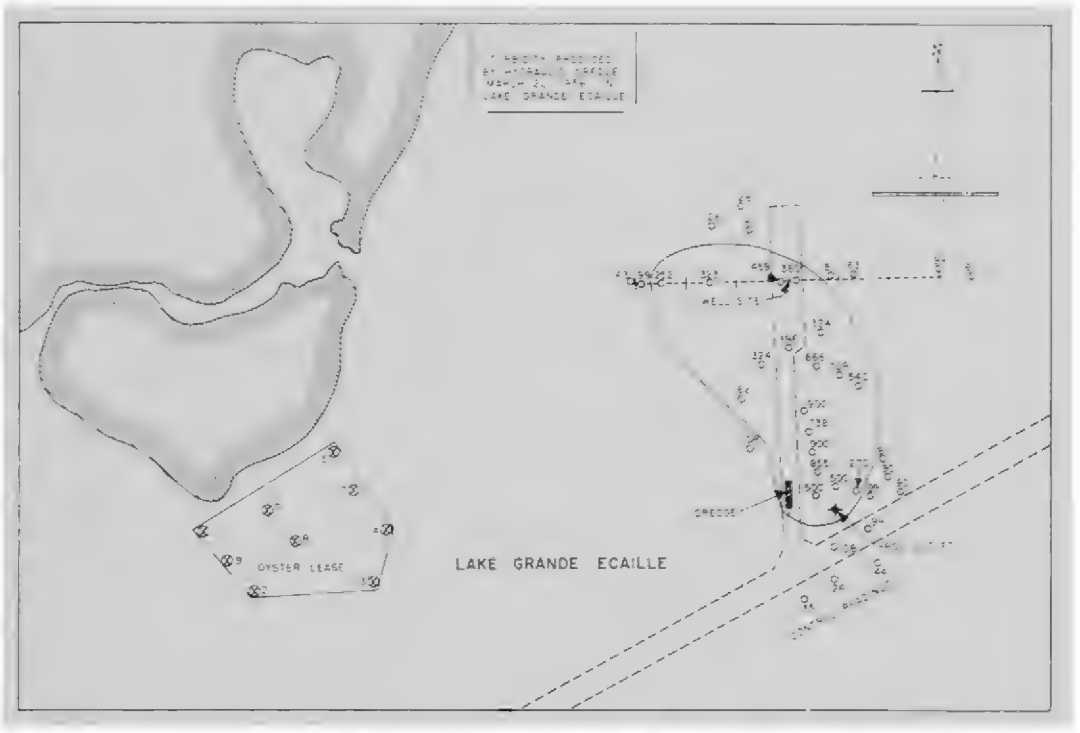

FIG. 11. Turbidity produced by a hydraulic dredge March 22, 1956, in Lake Grande Ecaille.

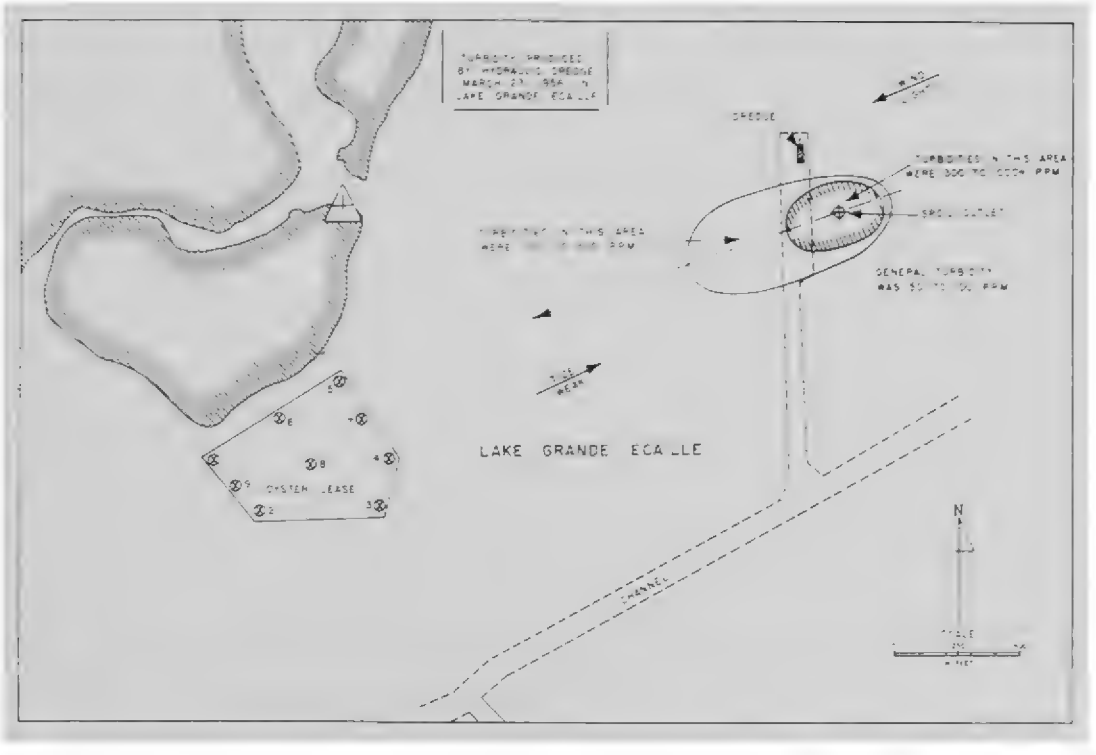

FIG. 12. Turbidity produced by a hydraulic dredge March 23, 1956, in Lake Grande Ecaille.

(Fig. 12). Currents were quite sluggish on this date. The spoil outlet was directed to the west of northwest and turbid materials were moving against current for about 200 feet, then drifting around and past the discharge.

Calculations based on samples taken along the axis of the turbid drift on March 22 showed that the mean decrease in suspended matter away from the discharge pipe was about 92 ppm per 100 feet (Fig. 13). Close to the discharge it was greater than that and with distance the rate of settling decreased. Part of the decrease in turbidity with distance away from the dredge was not due to fall-out of suspended materials, but to dilution with distance from the dredge discharge. The turbid area widened from the point of discharge. On March 22, the area of increased turbidity reached a width of about 700 feet at about 1000 feet from the dredge outlet. Under these conditions dilution alone reduced turbidities at 1000 feet to less than 100 ppm where the mean turbidity within 100 feet of the discharge was 1000 ppm. These data suggest that, except for the spoil dumped more or less directly on the bottom, the fines which drifted away were lost in the natural turbidities at distances above 1000 feet, and had the same effect on the environment as the materials put in suspension by natural agencies. Dilution may be a more important reason for decrease in turbidity with distance from the dredge than deposition on the bottom, a factor not taken into account in previous studies.

In this operation if we assume that the turbid materials within 100 feet of the cutterhead belong to the spoil, the relative amount dispersed can be roughly calculated. Roughly 19,000 cubic yards were dredged if the excavation was four feet below the mud-water interface. With a specific gravity of 1.7 (established by weight to volume checks) this amount weighed about 55 million pounds. The amount drifting away from the spoil area was roughly calculated. The mean turbidity in the wedge-shaped turbid area observed on March 22 was about 500 ppm. The area of increased turbidity was a triangle roughly 700 feet across the base and 1200 feet long. The maximum current velocity was about 0.25 knot, indicating that the 500 ppm turbidity was renewed about 76 times in the total dredging period of roughly 30 hours. Based on these figures the total weight (and consequently, the total volume) of material which drifted away from the dredge was less than 0.1 percent of the total removed in the digging operation. This rough calculation makes no claim of accuracy, but tests have been repeated several times with results varying up to 1.0 percent. Apparently in an ordinary dredging operation the amount of fines drifting away is roughly of the order of a maximum of one percent of the total materials removed from the bottom, an important approximation

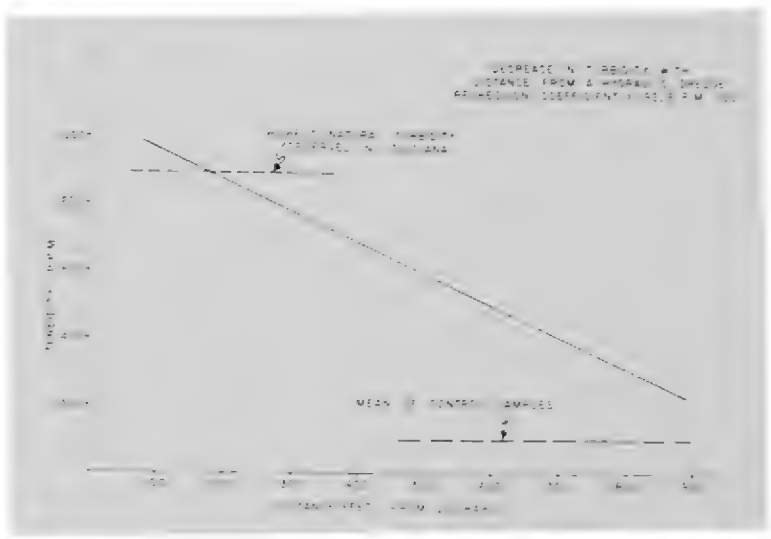

FIG. 13. Decrease in turbidity with distance from the outlet of a hydraulic dredge.

for considering the possibilities of silting of oyster beds. As an example, the oyster bed lying to the west of the dredging area in Fig. 11 is about seven acres in area. If all of the fines which drifted away settled on this bed (and none on the intervening bottom), the deposit would have averaged 0.2 inch thick on the bed, assuming a maximum drift away of 1.0 percent of the spoil.

Effects of High Turbidities on Oysters

Studies in the period 1947 to 1950 showed that in the field there was an inverse relationship between high turbidity and mortality of oysters. Oysters in the lower part of Barataria Bay, where salinity was generally high and turbidities relatively low, had consistently heavy mortality. In the fresher waters above Barataria Bay turbidity was higher (but not much higher). There oysters had only 1/3 to 1/2 of the mortality rates which occurred in the lower bay. Failure to find any kind of relationship between mortality and high turbidities caused a lack of interest in these matters.

However, in 1955, the effect of suspensions of mud on feeding and survival of oysters was tested (Mackin, 1956). Concentrations of mud from 100 ppm to 700 ppm were used and an apparatus was developed to keep the mud in suspension indefinitely. For mechanical details refer to the original report.

The data from that series of studies are condensed in Tables 7 and 8. Table 7 contains the data on survival and Table 8 the data on capacity to feed in high suspensions of mud.

These studies show no effect of the suspended mud. Oysters fed in suspensions up to 700 ppm without any apparent disadvantage, and mortality was on the average almost identical in control and experimental aquaria. It seems reasonable to assume that oysters would, with some unknown heavy concentration of suspended mud, lose the capacity to function normally. Field data showed that this heavy concentration could only be found within the area of direct spoil deposit by a dredge, where bottoms were covered. It was apparently impossible to maintain a suspension of high concentration long enough to cause mortality of oysters.

TABLE 7

Comparison of amounts of suspended mud cleared from experimental and control aquaria by ciliary action of the oysters with various concentrations of mud

Experiment no.	Turbidity; experimental tank, ppm	Mortality; experimental tank, percent	Turbidity; control tank, ppm	Mortality; control tank, percent	Dates
1	100	0	15	5	2–28–55 to 3–21–55
2	200	15	15	20	4–5–55 to 4–26–55
3	300	10			
4	400	35	10	25	4–27–55 to 5–18–55
5	500	20			
6	590	15	15	15	5–23–55 to 6–12–55
7	710	20			

TABLE 8

Data from experiments testing the effect on oysters of graded turbidities produced by mud suspensions. (20 oysters used in each tank)

Experiment no.	Experimental aquaria			Control aquaria			Percent cleared	
	No. samples compared	Mean turbidity at aquaria inlet, ppm	Mean turbidity in aquaria, ppm	No. samples compared	Mean turbidity at aquaria inlet, ppm	Mean turbidity in aquaria, ppm	Experiment	Control
1	37	100	92	38	15	10	8.0	33.0
2	28	200	178	36	15	10	11.0	33.0
3	28	300	270				10.0	
4	36	400	358	40	10	4	10.3	60.0
5	35	500	460				8.8	
6	28	590	541	35	15	4	8.3	73.3
7	30	710	630				11.3	

Canal Dredging and Dissolved Oxygen

Although there is little literature bearing on the matter, there have been complaints that when a canal is dredged, the spoil exerts an oxygen demand so strong that it depletes the oxygen in surrounding waters, thus killing oysters in the immediate area of the spoil. According to this theory a cycle is set up wherein the first oysters dead of asphyxiation then deplete the oxygen for greater distances.

An illustration of this type of claim was studied in Manilla Bayou, located northwest of Barataria Bay. An access canal was dredged to a well location in that area. This canal crossed Manilla Bayou in which was located an oyster lease. The bayou was quite deep (for the region), about 16 to 20 feet, and was bottomed with hard packed clay. Here and there in the deep bottom of the bayou were natural growths of rather thinly scattered but fairly good oysters. A strong scour current ran through the part of the bayou containing the lease (Jung, 1957). Salinities were generally quite low.

In crossing the bayou, the dredge did not remove spoil in the leased area, excepting that a section of a shallow mud flat on the west side was removed. The natural oysters in this bayou all grew in the deep parts of the hard clay bottom.

The claim was that all oysters were lost on five acres of the lease, 2000 sacks in all. The claim was supported by reports by Mr. Percy Viosca (no date) and Bruce H. Strawbridge (1956) made after an investigation on May 17, 1956.

This case came to trial on the 24th of June, 1958. After hearing testimony for the plaintiff the presiding judge directed the jury to return a verdict in favor of the oil company. The case itself was of relative unimportance, but the type of the claim is of considerable interest.

It was advocated in the lawsuit that the dredge dumped sufficient mud on the oysters in the area where the canal crossed the bayou to smother some of them. Since the area involved in this alleged direct covering could account for only a few sacks of oysters at most, this was considered only in the nature of a catalyst. It was stated that the oysters which were covered disintegrated and in doing so depleted the oxygen in the vicinity. This killed more oysters, which is turn disintegrated, in turn killing more. It was

testified in substantiation of this theory that a dead oyster utilized 50 to 100 times as much oxygen as did a live one and that the chain reaction set up as described could have extended over a number of days. It was claimed also that organic materials in the spoil bank had a strong oxygen demand and presumably enhanced the effect of the disintegrating oysters in reduction of oxygen.

Biochemical Oxygen Demand of Disintegrating Oysters

There are data at hand to calculate roughly the order of magnitude of the B.O.D. of market size oysters in disintegration.

Gaarder and Alvsaker (1941) made analyses of carbon content in *Ostrea edulis*. They found that carbon makes up about 48 to 50 percent of the dry weight of the meats. It is assumed that the amount of carbon in *Crassostrea virginica* is about the same.

In the laboratory at Grand Isle the author has determined the dry weight of many oysters. This was done by means of the drying oven, and is not an exact method, the actual dry weight being less than found by this technique. Calculations based on measured dry weights are conservative. The mean dry weight of more than 200 market-size oysters was 1.7 grams.

Since the ratio of carbon to oxygen in carbon dioxide is 1 to 2.66 approximately, the amount of oxygen needed to oxidize the carbon in an oyster is 2.66 times the weight of the carbon. At temperatures ranging from 20°C to 26°C, oysters of market size disintegrate in 5 to 7 days (Mackin, 1954). Using these data, the amount of oxygen used in disintegration can be roughly calculated to be about 2.26 grams per oyster per six day period, or about 16 mg per hour.

This should be compared with respiration requirements of living oysters. Collier (1959) has found that oysters may use up to 50 mg of oxygen per hour when pumping water at a rate of 25 liters per hour. He found that the oxygen consumed varied with the pumping rate, and was greater in small oysters than in larger ones. He stressed that oxygen uptake measured under different conditions cannot be properly compared.

Galtsoff and Whipple (1931) found that oxygen consumption of oysters varied from 6.45 to 15.04 cc per hour (760 mm Hg, 0°C) per 10 grams of dry weight. This rate is equivalent to 1.5 to 3.5 mg per hour per oyster. Later Galtsoff (1947) used improved techniques and found that adult oysters consumed 1.57 to 8.29 mg per hour at 25°C. These were starved oysters and the studies were designed to show the oxygen uptake at basal metabolism level. Galtsoff pointed out that he avoided any condition, such as shell movement or feeding, which would increase uptake of oxygen. His figures probably represent the minimum requirements.

Galtsoff's figures indicate that the oysters respired about 5 mg per hour at 25°C when using least oxygen. By comparing the 5 mg per hour with our calculated figure of 16 mg per hour B.O.D. in disintegrating oysters, it is clear that dead oysters do not use 50 to 100 times as much oxygen in disintegration as do live oysters.

Galtsoff's figures indicate no significant difference in the amount used by dead and normally feeding live oysters, and Collier's work (1959) indicates that live oysters while pumping may actually use more oxygen than do dead oysters.

Sparks, Boswell, and Mackin (1958) studied the comparative capacity of dead and live oysters to reduce oxygen content in aquaria using a B.O.D. method. Live and dead oysters of the same weight were paired and the reduction of oxygen in the containers

(jars, battery jars, and aquaria) was measured over various periods of from one-half hour to 192 hours. Data were taken on 35 pairs of live and dead oysters along with controls (blanks). The live oysters reduced oxygen an average of 3.25 ppm, and the dead oysters an average of 3.51 ppm. These experiments confirmed the conclusion already arrived at by calculation: there is no significant difference in the rate of utilization of oxygen by live and dead oysters. This being true, it is not possible that dead oysters on natural bottoms use such large amounts of oxygen that they deplete the water in their vicinity. Also, the oxygen requirements of the dead oysters come to an end in 5 days to a week, while that of the live oysters continues.

There are some practical considerations to be mentioned in this connection. The matter of comparative utilization of oxygen by live and dead oysters is to some extent academic. Oysters rarely disintegrate when they die under natural conditions. Crabs and fishes destroy them long before that happens. Through 12 years of studies, our greatest problem has been the recovery of sufficient gapers (nearly dead oysters, with shell open) for significant study and to attain this end cages for exclusion of scavengers have been developed. Even with the best of equipment and frequent visits, only 33 percent have been recovered in the best record attained in the field (Mackin and Sparks, 1958). Laboratory recovery is better, where oysters may be examined once or twice a day.

This problem may be approached in a different manner. Assuming that a disintegrating oyster uses 16 mg of oxygen per hour, what percent of the total oxygen available can dead oysters remove at minimum current conditions? In Manilla Bayou, it was claimed that oysters on five acres in the amount of 2000 sacks with 208 oysters per sack were killed by asphyxiation. At 16 mg per hour each this number of oysters would require about 6.7 million mg of oxygen.

Jung's (1957) study of currents showed that at the equatorial tide period the minimum flow in Manilla Bayou was 366 cubic feet per second (at maximum wind and tide it was 2537 cubic feet per second). Normal oxygen level has been established by Jensen (1949) who took 1132 samples for oxygen titration in Barataria Bay. The oxygen content was established as being in excess of 5.00 ppm for most of the time. Using 5 ppm as a basic figure it was calculated that the minimum amount of oxygen passing through the bayou was about 408 lbs per hour, or about 185 million mg. The 2000 sacks of oysters were able to reduce the available supply of oxygen only about 3 to 4 percent.

Manilla Bayou has a strong scour current and flushes in a matter of hours, and the above calculations do not take into consideration the capacity of natural waters to produce oxygen by photosynthesis or other means.

Oxygen Demand of Spoil

Is it true that the "anaerobic muds," which are brought to the surface by a dredge, have a greater oxygen demand than do the muds at the mud-water interface of undisturbed bottom? This question may be answered by determining whether the oxygen demand of bottom muds increases or decreases with depth below the surface.

Waksman and Hotchkiss (1937) measured B.O.D. of marine muds at different levels. They found that the top centimeter of muds from cores had significantly higher oxygen demand than did muds at greater depths. Anderson (1940) determined the organic content of many marine muds and found that the highest organic content and the greatest B.O.D. was in the top of his cores. This author showed also that in a period of 15 days,

only 2 to 7 percent of the carbon present in mud is available for bacterial oxidation. Waksman and Hotchkiss stated that "the organic matter itself was not absolutely inert, but could be gradually, even if only very slowly, decomposed."

Moberg *et al* (1937) also showed that the highest organic content of marine muds is at the surface of the cores. Trask (1939) found that there was about a 15 percent decrease in organic matter in 30 centimeters of depth in marine sediments. Trask found that only anaerobes could alter long-buried organic materials.

The authors referred to are in accord that (1) oxidation of organic substances in marine muds is a very slow process, (2) only a small fraction of the total carbon present can be attacked in 5 to 15 days, and (3) both the amount of organic materials and the rate at which it is attacked is reduced with depth. This may be due to the nature of organics stored in muds, being largely of cellulosic nature and lignin. It seems clear that any rapidly oxidizable substances are destroyed before plants are buried and incorporated in the muds. Anaerobic decomposition continues, at a slower rate, after burial.

There are, however, other substances in muds which have an oxygen demand. Hydrogen sulphide generated bacterially in the muds may be oxidized, and may create a demand in excess of the organic demand. The oxygen demand of this gas is immediate and being a gas it must be oxidized immediately or be lost to the atmosphere. Hydrogen sulphide can have an effect on oysters or oxygen only if liberated in considerable quantity over long periods of time. All natural mud bottoms produce some H_2S and in small enclosed ponds may materially reduce oxygen during hours of darkness.

Sudden liberation of large quantities of hydrogen sulphide by a dredging operation may not do more than reduce oxygen for a few hours. Since oysters are resistant to complete oxygen depletion for several days to a week (Von Brand, 1946), a temporary oxygen reduction could not be effective in producing mortality.

Gases emanating from deep marsh muds in Louisiana are common. Analyses of samples of these gases (Jensen, in Seiling, 1951) show that generally the methane content was from 19 to 87 percent. Common atmospheric gases were present in smaller amounts, but hydrogen sulphide was not found in the samples studied. The indications are that hydrogen sulphide forms more commonly in surface muds and is either (1) liberated as formed, or (2) stored in the muds as metallic sulphides, or may even progress to pyrites. Pyrites are common in woody materials formed in the bottom muds. Small nodules of metallic sulphides are often found in foraminiferan shells. Oyster shell which has been long buried takes on a greyish to black color due to metallic sulphides which form in the shell. Treatment with hydrochloric acid will liberate hydrogen sulphide rapidly, or the sulphides may be consumed more slowly by oxygenation in air.

Methane formed in large amounts in bottom muds ordinarily bubbles through water and is liberated to the atmosphere. It is almost insoluble in water, and is quite inert chemically. Large quantities of methane (in natural gas) bubbled through trays of oysters for months at a time failed to produce mortality (see Menzel, 1949).

Study of Sediments in the Vicinity of Dredging Operations in Lake Grande Ecaille

In this section will be presented data from a study made in the fall of 1955 and spring of 1956 in connection with two claims that damage to oyster beds or oysters resulted from deposits of dredging silt.

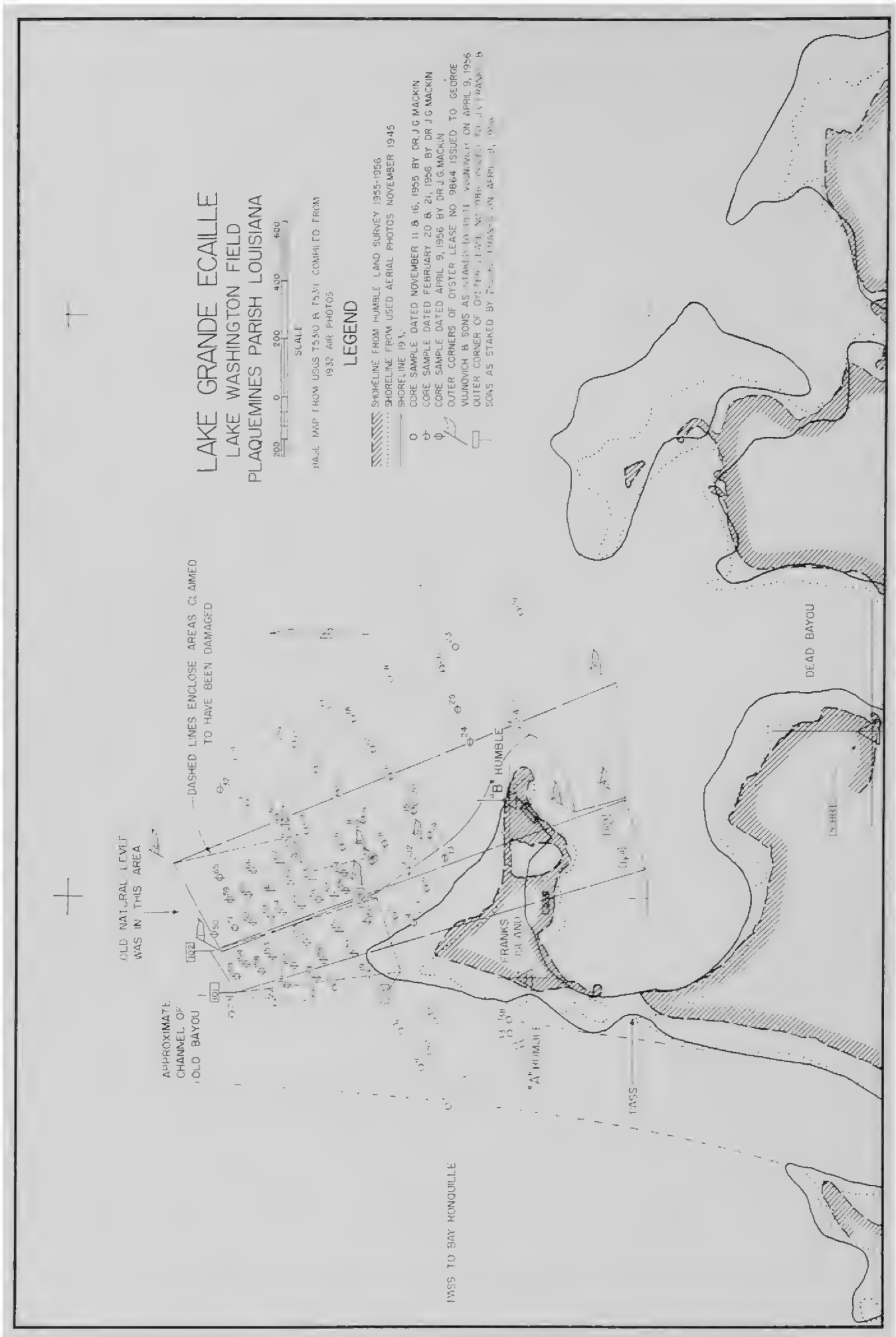
LAKE GRANDE ECAILLE
LAKE WASHINGTON FIELD
PLAQUEMINES PARISH LOUISIANA
SCALE
LEGEND
SHORELINE FROM HUMBLE LAND SURVEY 1955-1956
SHORELINE FROM USED AERIAL PHOTOS NOVEMBER 1945
CORE SAMPLE DATED NOVEMBER 11 & 16, 1955 BY DR. J G MACKIN
CORE SAMPLE DATED FEBRUARY 20 & 21, 1956 BY DR J G MACKIN
CORE SAMPLE DATED APRIL 9, 1956 BY DR J G MACKIN
OUTER CORNERS OF OYSTER LEASE NO 9864 ISSUED TO GEORGE
OUTER CORNER OF
SONS AS STAKED BY
DEAD BAYOU
FRANKS ISLAND
OLD NATURAL LEVEE WAS IN THIS AREA
APPROXIMATE CHANNEL OF OLD BAYOU
PASS TO BAY HONQUILLE
PASS

FIG. 14. Map showing locations of cores taken on and around leases 9864 and 9861. The locations of these cores were surveyed and mapped by engineers of the Humble Oil and Refining Company.

The leases involved, numbers 9861 and 9864, were located in the southwest part of Lake Grande Ecaille (Fig. 14), straddling a small marsh island (Franks Island), on which were several oystermen's camps and a private fishing camp. Silt was said to have been deposited on a part of the north portions of these leases. It was believed by the owners of these leases that the Humble Company, "beginning about November, 1954

and continuing until about February, 1955, was engaged in dredging operations in Lake Grande Ecaille which resulted in dumping of spoils, sands, and other refuse material into the waters of Lake Washington or Lake Grande Ecaille." The spoils, etc., were, according to the lease owners carried by currents to the lease. There were a few oysters on one lease (9864).

Dredging Operations

Investigation showed that The Humble Oil and Refining Company had made extensive dredgings in Lake Grande Ecaille, before, during, and after the period from November, 1954 to February, 1955. Those dredgings are indicated on the map, Fig. 6, although not all of the canals shown were dredged in the period indicated in petitions of plaintiffs. Seventy-one thousand cubic yards of materials were dredged between May 17 and July 6, 1954, and the spoils were placed in spoil islands. The canal at the extreme east part of the bay was dredged in this period (see earlier section of this report), and the approximate location of the spoil island is shown on the map. Also the first and second access canals in the network of canals shown in the northeast area were suction dredged in this period and the spoil placed in a spoil island which is also shown. An additional 33,000 cubic yards was placed in spoil banks prior to November, 1954.

About 42,000 cubic yards of material were dredged after February, 1955, and spread on the lake bottom. This spoil was from the canals in the far northeast part of the Humble field.

In the period from November, 1954 to February, 1955, 452,000 cubic yards of bottom were dredged. The canals were all in the system of canals of the northeast part of Lake Grande Ecaille and the single long northwest-southeast canal to wells 28 and 29 which traverses the center of the lake. This latter canal was closest to the beds of plaintiffs. Of the 452,600 cubic yards dredged in the period named in the suits, 206,800 cubic yards were pumped into the marsh. Spoil from the canals bordering the marsh in northeast Lake Grande Ecaille was disposed of in this way except that from the tier of canals farthest to the north. Most spoil from those canals running westward from the central north-south axis of the northeast field was spread in a six-inch layer on the lake bottom, as was the spoil from the long canal to wells 28 and 29. The total thus disposed of was 245,800 cubic yards. The closest point of any of the dredging operations (the south end of the canal to well 28) was almost exactly one statute mile from the two leases. The farthest point away was a little over two miles.

Currents in Lake Grande Ecaille are mostly variable and move with the wind. However, the major inlet into the bay is Rattlesnake Bayou which empties into Lake Grande Ecaille to the northeast of the Humble field. Movement of water is from the northeast, wind disregarded, and leaves the bay by the pass just to the west of the two oyster beds. On ebb tide, therefore, what current there was moved generally from the canals in the direction of the beds, reversing on the flood. Least tidal current was in the center of the bay, which was also the deepest part of the bay.

Test Calculations

Assuming that one percent of the spoil (see Section IV) from the canals floated away with the current, the maximum mean depth of silt deposition possible can be roughly

calculated by making certain reasonable assumptions.

One percent of 245,800 cubic yards is 2,458 cubic yards of spoil which may have drifted away from the dredge outlet. This would be spread both to the northeast and the southwest, somewhat more than one-half moving to the southwest (because of the longer ebb period). It was estimated that the movement would be in about the proportion of 13.4 to 11.5. This would mean that 1,323 cubic yards was spread to the southwest, toward the two leases. Ignoring the fact that most of it would settle in the deepest part of Lake Grande Ecaille where current was least, it is assumed that this 1,323 cubic yards was spread over the approximate square nautical mile between the nearest canal and the beds (including the beds). If this happened the maximum thickness of the layer was 0.012 inch. If *all* of the one percent of spoil should in some manner move across the intervening one mile and all of it was deposited on the twenty-two acres of the oyster beds, the layer would have been less than one inch thick.

The theory that a drift of spoil from the dredges moved across the intervening mile and was deposited on the bed was rejected at once, since the rough calculation indicated that no considerable amount of spoil could have traversed the intervening mile to the beds. Even gross error in any part of the calculation or the assumptions would not alter the figures materially. There remained the possibility that the dredgings in some manner caused precipitation of suspended materials on the leases.

If the leases had been damaged by the dredging operations, it seemed certain that the intervening bottoms would also have been silted. There are no mechanisms by which dredgings in the northeast part of the bay could produce silting conditions on leases 9861 and 9864 without affecting the intervening bottom. Considerable attention was given to this intervening bottom, between the canal to wells 28 and 29 (see map) and the leases in question.

Characteristics of Lake Grande Ecaille

Lake Grande Ecaille is a roughly triangular bay with the acute angle pointing to the northeast and the base at the south. The most important outlet to the northeast is Rattlesnake Bayou. When the tide ebbs the water from the Lake and Rattlesnake Bayou converges to the southwest corner and passes into Bay Ronquille by the northern Four Bayous Pass. The entire area is roughly about 3.75 square nautical miles. U. S. Coast and Geodetic Survey charts give the depth over most of the bay as about 3 feet at mean low tide. It is deeper than that at present; over most of the lake it is at least four feet deep at mean low tide. On the date of January 10, 1956, the lowest low tide of the winter of 1955–56 occurred. On that date a speedboat was operated in Lake Grande Ecaille and the depth in the bay was measured at about three feet. Since the water on that date was more than a foot lower than mean low tide, it is clear that the depth of the lake must average more than four feet, as shown in measurements presented in Section I.

The bottom of most of the lake is soft mud which contains shells of small mollusca and foraminifera in greater or less concentration. Along the margins, especially the south shore, are areas with sufficiently stiff bottom that oysters may be fattened thereon. In most areas, these marginal bottoms are kept clear of soft silt accumulations by wave-wash and by along-shore tidal movement of the water, especially on the south shores.

Currents are generally quite sluggish in Lake Grande Ecaille. If we calculate the volume of water movement on ebb tide in Grande Ecaille as two-thirds that passing out

Quatre Bayou Pass (1005 million cubic feet, Fig. 1, Marmer, 1948) the maximum tidal current at transect a-b (see map, Fig. 6) is 0.25 knot. Currents increase when the constricted pass into Bay Ronquille is reached. There is thus an abrupt increase in water movement, just to the west of Franks' Island.

Lake Grande Ecaille was originally two or more isolated marsh lakes. One of these was in the northeast area, and another in the south to southeast area. The west end of Lake Grande Ecaille was once completely closed off by the natural levee of the bayou which lies to the immediate west of Franks' Island. The northern part of this bayou (Figs. 1, 6, and 14) can be clearly seen continuing into the marsh to the northwest of the bay. At some time in the past this levee was broken through as subsidence, rise in Gulf level, and erosion reduced the land. Subsequently, the break-through widened until now it is about a mile across. Franks' Island is a southern remnant of this old natural levee. A study of the topography in that area shows that the reduction of the original natural levee probably took place by a series of successive breaks through the levee, each isolating a point of land as an island, as is Franks' Island now, the last to be so isolated. These breaks divert the current to the south of the island. Erosion by sinking and wave action then destroys the island so formed, another break-through occurs to the south to form another island and the process is repeated.

The south shore of Lake Grande Ecaille is particularly subject to erosion. The map, Fig. 14, shows this fact clearly. It is calculated that East Island, which lies to the east of Franks' Island, broke up at the average rate of about 60,000 cubic feet per year for the last 24 years. It is now represented by only a small remnant of marsh. (Since this was written, East Island has disappeared completely.) According to Coast and Geodetic Survey charts, the area of the island in 1932 was about 9 acres. Franks' Island had a similar rate of erosion until shells placed around the island by oystermen arrested the erosional process in about 1945. The north shore of Franks' Island receded by more than 100 feet in the period from 1932 to 1945 (see map).

Lake Grande Ecaille long ago ceased to be a natural oyster producing area. How long ago that was is not known, but the complete absence of any natural oyster reefs either dead or alive means that it was a long time. Within the last twenty years or more it has been used more or less successfully as a winter fattening area. At present its value depends entirely on winter fattening. Conchs and heavy fungus disease make it almost impossible to keep oysters on the beds in summer. Oystermen have largely abandoned Lake Grande Ecaille.

The Area of Leases 9861 and 9864

These leases extend across Franks' Island, part of the leases lying to the south and the remainder to the north (Fig. 14). That portion of the leases which lies to the north of Franks' Island is of most importance. A study of the bottom, sediments, etc., shows that the area to the north of Franks' Island was itself originally an island which was reduced by erosion. The *in situ* sediments of the original island are now identifiable because there are no shells of marine animals found in them. A core passing through the recent marine sediments encounters the marsh-laid sediments of the old island only six or eight inches down over most of the area. This old island was originally separated from Franks' Island by a channel. Before the pass to the south of Franks' Island was opened up, the current passed across to the north of the island (as part of it still does), following this old channel.

The leases are on a slightly raised part of the bottom and slope off to the west into Bay Ronquille Pass and also to the north and to the east. The highest part is an artificially built up area (Fig. 15) composed of *Rangia* shell.

On February 21, 1956, at about 1400, a series of soundings was made in the area of the leases. A Subscout sounding device was used and checked against direct pole readings. Six parallel lines of soundings were made in a north-south direction, beginning in each case about fifty feet from the shoreline of Franks' Island. The depth was found to vary from 5.2 feet to 5.9 feet over the area of the leases. Corrected to mean tide level, the depths were 4.3 feet to 5.0 feet, the shallowest point being on the artificial *Rangia* shell reef and sloping away in all directions. Error here is probably about ± 0.3 foot. Assuming this much error, the depths shown are significantly greater than those shown on Coast and Geodetic Survey navigation charts. Data on which these charts are based were taken in the decade from 1930 to 1940.

Microfaunal Analysis

Careful examination of the cores taken in the area of study provided evidence of the origin of the sediments. Foraminiferan shells were retained in sieves varying from 1/200 to 1/40 inch mesh size. Shells of molluscs, sand and woody debris were found distributed throughout all the sieves.

Foraminiferan shells were found in all cores taken in Louisiana which were composed of marine muds deposited at a normal rate of speed. Because most bottoms are in equilibrium, or depositing rates are slow, foraminiferan shells accumulate in the surface layers.

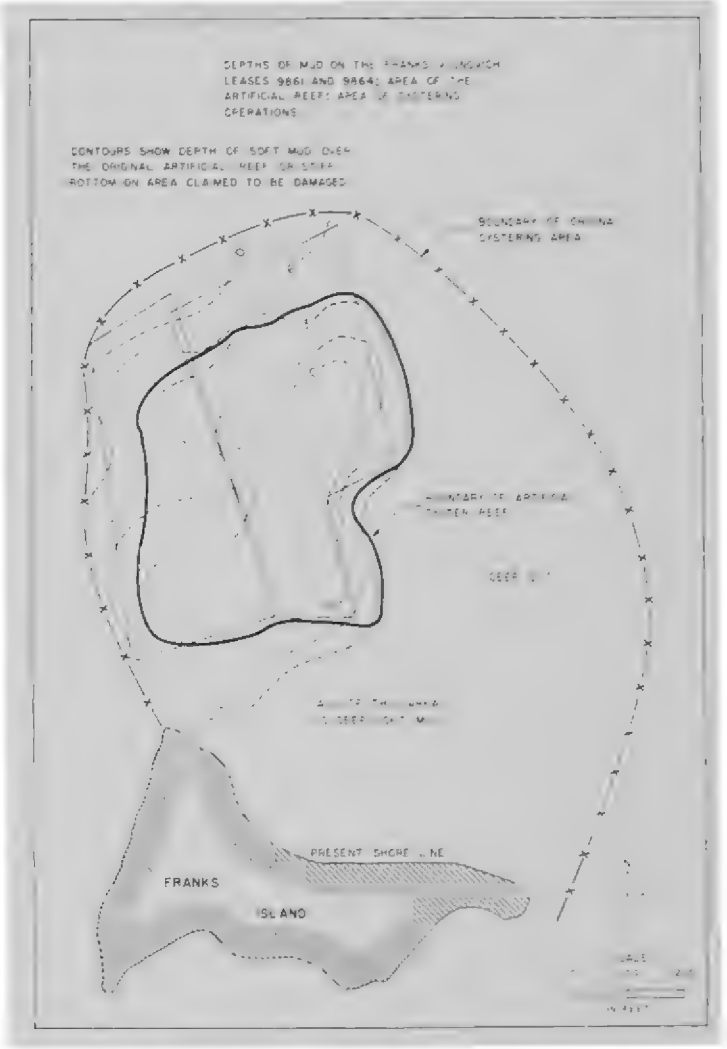
CONTOURS SHOW DEPTH OF SOFT MUD OVER
THE ORIGINAL ARTIFICIAL REEF OR STIFF
BOTTOM ON AREA CLAIMED TO BE DAMAGED
BOUNDARY OF OYSTERING AREA
PRESENT SHORE LINE
FRANKS
ISLAND

Fig. 15. Depth contours of silt; the artificial reef and core locations on the alleged damaged area of the Franks-Vujnovich leases, Lake Grande Ecaille.

These layers may be slowly covered with later deposits, and become buried more or less deeply. However, in areas where slow scour is taking place, winnowing may uncover successive layers of foram shells which are concentrated in and near the mud surfaces.

Abnormally rapid deposit, such as was claimed in the case of leases 9861 and 9864 may precipitate suspended silts on the bottom so rapidly that foraminiferan accumulation would not be possible. Such rapidly deposited layers do not contain forams, nor do they contain shells of molluscs, large woody debris or any other particles too large and heavy to be moved by means of ordinary current.

Figs. 16 and 17 show different types of foram shells. The spiral forms are most common, especially those with calcareous shells. These may contain small nodules. These are mostly metallic sulphide concretions. Spirals with chitinoid shells are common in muds in sluggish bayous, where the calcareous shells are absent. It seems probable that some highly reducing muds may destroy the calcareous shells.

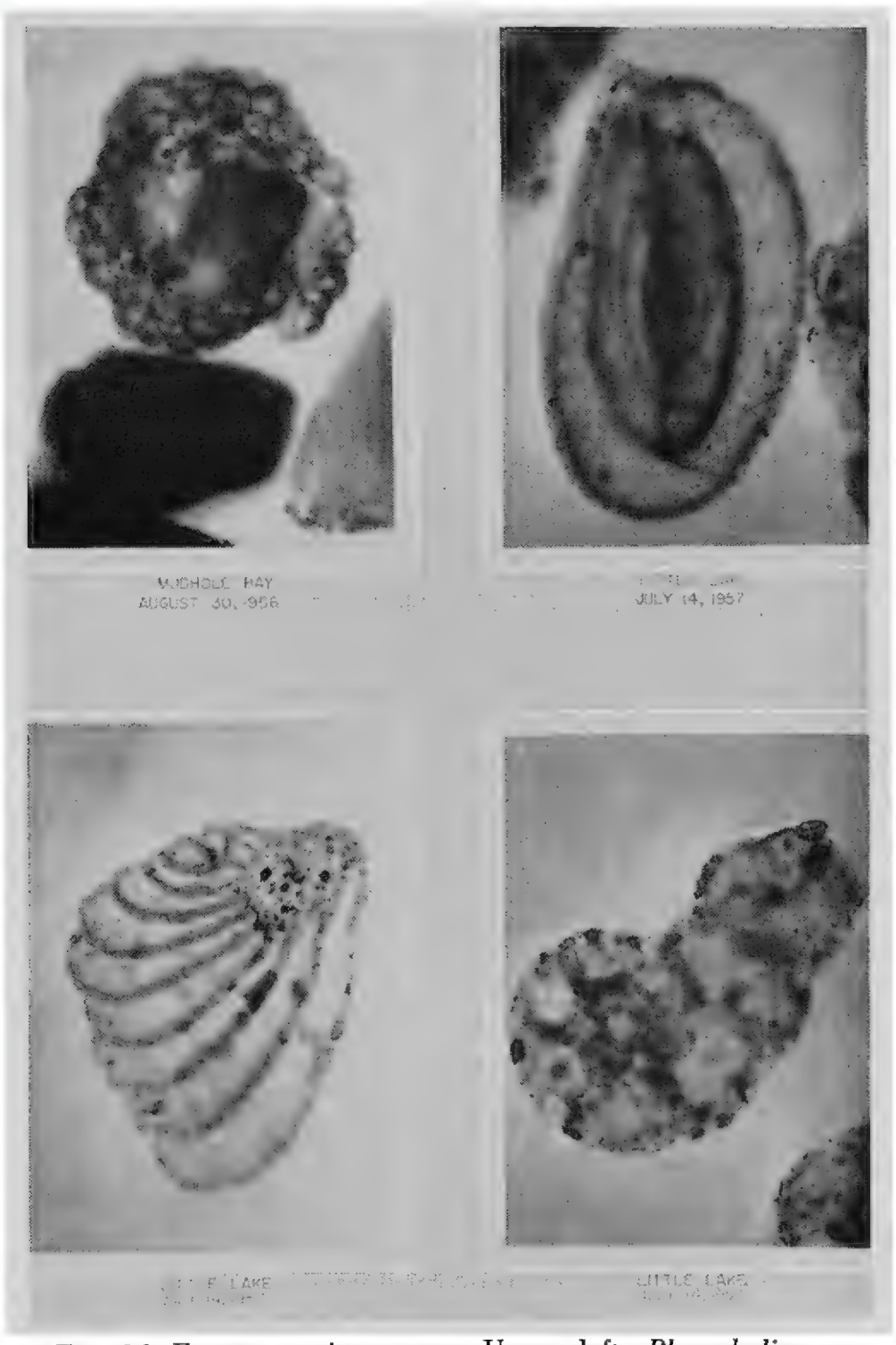

FIG. 16. Forams, various types. Upper left, *Planorbulina sp.;* upper right, *Milliammina fusca;* lower left, *Ammoastuta inepta:* lower right, *Ammobaculites sp.*

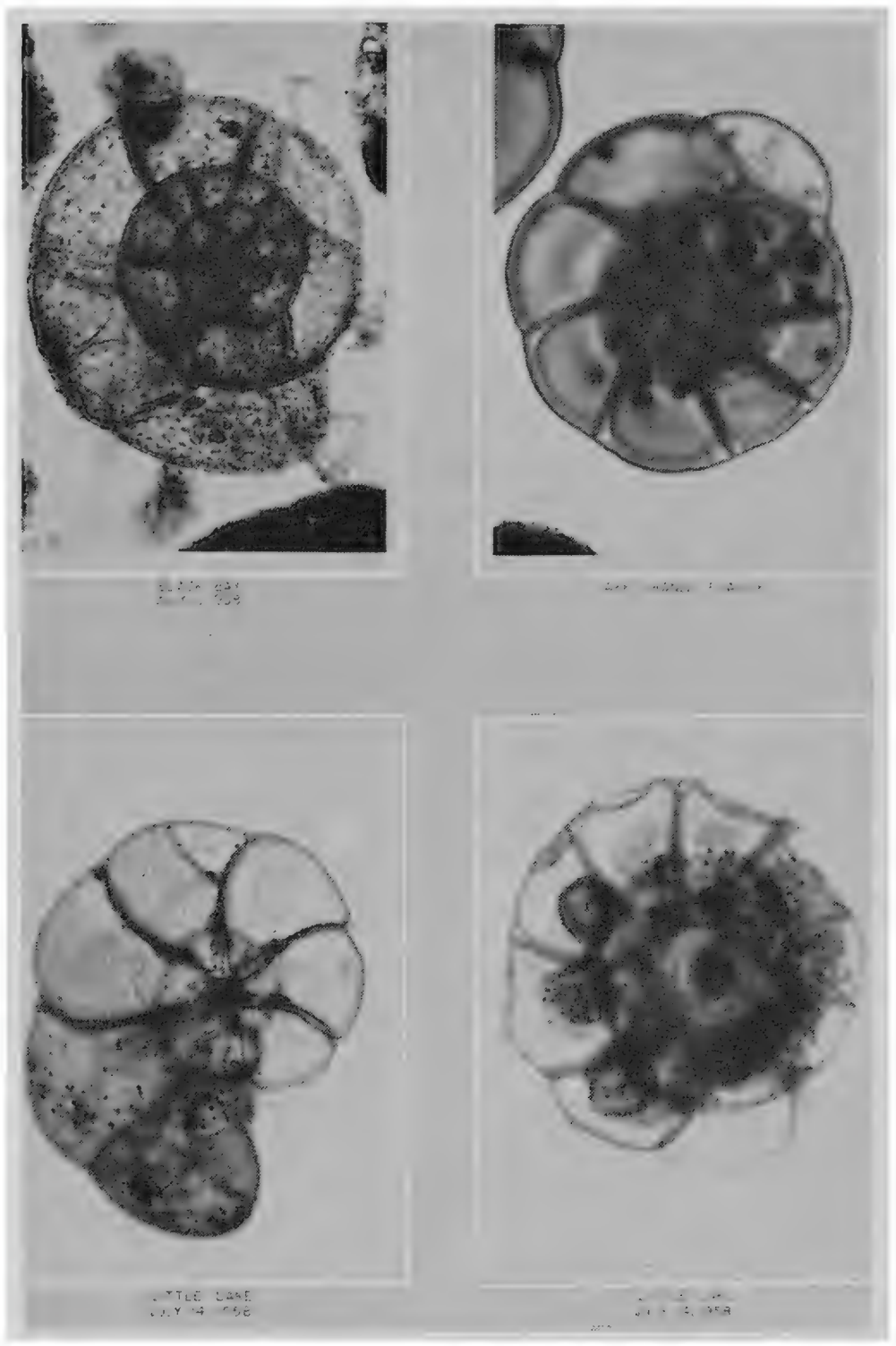

FIG. 17. Forams, spiral types. Upper left, *Trochammina sp.;* upper right, *Streblus beccarii;* lower left, *Elphidium poeganum;* lower right, *Streblus sp.*

Forams which produce the "sand grain" cases are common in low salinity areas, and are present in smaller numbers in higher salinities. Some of these are roughly spiral and some are flattened and oblong. In Fig. 16 is shown one of the chitinoid forms found commonly in marsh ponds. These are so characteristic of the marshes that they rarely are found far from shore. They have been taken in great abundance in marshes where no ponds are presently found. There are several different species of these interesting forams which look oddly like an insect wing under the microscope. The oblong shell shown in Fig. 16 appears not to be confined to any single habitat, but is much more abundant in bayous of low salinity than in open bays.

Shell fragments from muds are shown in the photographs, Figs. 18 and 19. The shell in Fig. 18 is blackened with the sulphide characteristic of long-buried fragments of oyster shell. There are some intermixed shells of other molluscs, which retain a white color irrespective of length of time of burial. In Fig. 19 the shell is nearly all white,

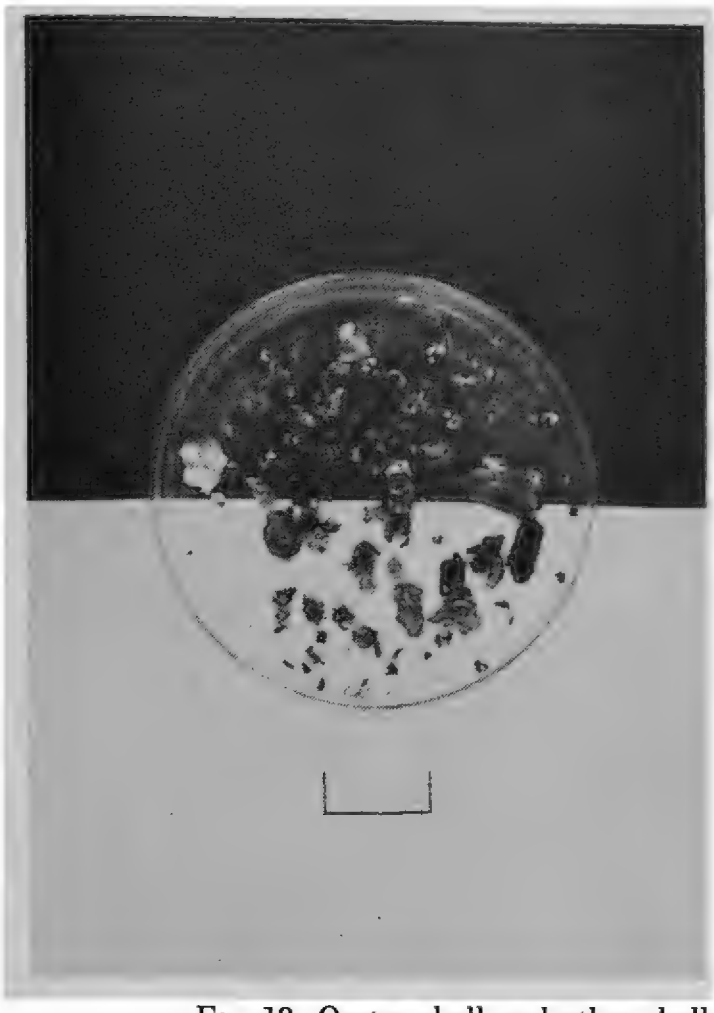

FIG. 18. Oyster shell and other shell from the Franks-Vujnovich leases. (Left)

FIG. 19. Shell fragments from a core in Lake Grande Ecaille. (Right)

showing against a black background. The plant debris showing over the white background is characteristic of marsh muds.

Origins of silt may sometimes be detected on the basis of reduction of weight of muds with ignition. Loss on ignition does not give an accurate measurement of organic content of the muds, but the measurement is comparative between samples. Since most carbon compounds in the muds are derived from marsh plants, highest concentrations are marginal in the bays and decrease toward the centers (see Mackin and Hopkins, 1961, Section 5). Establishment of a decrease in values of loss by ignition, from the shore to offshore areas, would indicate that silts on leases 9861 and 9864 were largely derived from disintegrating and slumping marsh shorelines.

When leases 9861 and 9864 were first studied it was found that they were, for the most part, quite silty and soft. Muds were found by probing to be almost mushy in places, while others were quite hard and with the consistency of a shell reef. No natural reef was found. All gradations of depth of soft silt were found up to more than one foot and all gradations of consistency from very hard to very soft were observed. The problem resolved itself into determining whether or not the Humble Company dredging operations were responsible for the soft silts found on some portions of the bed or whether the observed silts were the result of other factors.

CORES FROM THE MUDS ACCUMULATING IN THE CANAL DREDGED TO WELLS 28 AND 29

It was indicated above that the access canal to wells 28 and 29 was dredged within the period from November, 1954 to February, 1955, when it was claimed that leases

9861 and 9864 were silted. It was also the closest canal to leases 9861 and 9864. Wells 28 and 29 were dry holes and the canal was abandoned shortly after the drilling was completed. When the investigation of the claims began about a year after the dredging operation, the canal had silted up level with the surrounding bottom. Abandoned canals in low-current-velocity areas always silt up. While canals are in use, propeller wash may prevent silting to some extent.

The silt which was deposited in the canal was used to determine the characteristics of silts carried by currents in Lake Grande Ecaille. The numbers of foraminiferan shells and molluscan shell fragments, the sizes of woody fragments, and the loss of weight on ignition, were determined for cores taken in the canal, located approximately on the center line between the two margins. Engineers of the Humble Oil and Refining Company surveyed the canal and the locations of the cores were plotted (Fig. 6). Field examination of preliminary cores showed that they were composed of a uniform silt without any macroscopically visible shell, sand, or woody debris except at the bottom where the coring tube penetrated the original bottom. Two cores were taken at each location. These were analyzed microscopically. This analysis showed the following:

(1) Foraminifera were very rare. None were found in the silt in one core, only one in another, and ten in the third. All of these foram shells were very small, all being less than 1/80 inch in diameter.

(2) Shell fragments were also rare. Only one was found which was classed as large (about one-half inch). All others (12 total) were very fine fragments and all were flattened flakes. One core had no shell at all.

(3) Woody debris was comparatively small in amount, and was composed of very fine fragments. In only one core were woody fragments of any size found and there were only a few of these. However, examination of the silt which passed through all sieves showed that there was very fine plant debris in considerable quantity. In none of the cores were there layers of peaty materials such as is found in undisturbed bottom.

(4) Sand was present only in the fine sizes, no medium or coarse sand being present. Sand here means silica sand, but the sizes are according to the conventional geological classification. These data are summarized in Table 9.

Spoil from the original canal was spread over the bottom, the closest part of the direct depositing area being 50 feet from the canal. Since the canal itself was 65 feet wide, the cores were about 32.5 feet from the margins of the canal. It was obvious that water transport of forams, shell, and large woody debris was extremely limited even at the short distance involved. It is believed that the forams developed in the mud as it accumulated but there could have been limited transport. The canal was cut into the bottom to a depth of approximately four feet so that roughly the silt was deposited in the abandoned canal at a rate of about four inches per month, if the rate was uniform through

TABLE 9

Numbers of foraminiferan shells, and fragments of molluscan shell from three cores in water-borne silt fill in abandoned canal to wells 28 and 29

Core no.	No. of forams per inch of core	No. of shell fragments per inch of core
16	0.3	1.7
14	0.0	1.8
15	5.0	0.0

the year of deposit. The data indicate that forams may not live to reach mature size if the mean rate of deposit is about four inches per month.

Muds from the cores taken in the canal to wells 28 and 29 were dried to constant weight and then ignited in a muffle furnace. The results are presented in Table 10. The data show that the range of values is comparatively small in ten samples, the mean amount of 14.4 percent loss of weight is only moderate, and there is no difference in the ignitable materials in the top six inches and in mud more than six inches below the surface. These values will be compared with others in the table later.

Cores From the Area Northeast of the Dredging

Two cores were taken on February 20, 1956, in the area to the northeast of the canal to wells 28 and 29. These locations are shown on the map, Fig. 6. One (No. 397) was quite close to one of the more northerly canals, near well K, and another (No. 738) was just to the northeast of well 29. Each of these was 150 to 175 feet from the respective closest canals. Neither was in a spoil area. It was believed that cores taken in this area would show silting if any part of Lake Grande Ecaille had been affected by water-borne silt, or precipitation of silt, caused by the dredging operations.

Macroscopic study of these cores showed that there was about one inch of a light-colored soft mud on the surface. Below that, there were numerous shell fragments down to about six inches below the surface. This part of the cores was moderately compact. Below the six-inch level, the cores were composed of compacted clay-like mud and there were peaty layers in the bottom of the longest core. This one was twenty-one inches long. Shell was abundant in the bottom six inches of this core.

Microscopic study showed that the soft mud in the surface inch of both of these cores had 100 to 200 large forams and a small amount of shell. There was no indication that silting had occurred in the area where they were taken. It is certain that the bottom was normal below the top inch of soft materials. Since one-fourth to one inch of a soft flocculent material is common in cores from bay bottoms, it is probable that there had been no deposition even in the immediate vicinity of the canals. The presence of considerable numbers of forams and some shell in the top inch indicates that the difference in color of the top inch was due to presence of oxygen which prevents the formation of

Table 10

Loss of weight on ignition by various muds. Values are percentages of the dry weight of the samples

Location of samples	No. of samples	Mean loss of weight, percent	Range of loss of weight in these samples, percent	Distance from spoil deposit area
Silt trapped in canal after abandonment; top 6 inches of silt	4	14.4	11.9 to 17.4	50 feet
Silt trapped in canal after abandonment; below 6 inches	6	14.4	12.2 to 18.3	50 feet
From spoil area: in top 6 inches	12	8.3	4.0 to 12.3	Zero
From spoil area: below 6 inches (below spoil)	7	12.4	7.0 to 18.3	Zero
Bay bottom: top 6 inches	15	10.3	6.5 to 21.9	1500 to 2500 feet
Bay bottom: below 6 inches	4	27.7	16.7 to 40.1	1500 to 2500 feet
On leases of Franks-Vujnovich: top 6 inches	55	16.3	7.1 to 38.0	More than 5000 feet
On leases of Franks-Vujnovich: below 6 inches	26	33.4	6.4 to 67.2	More than 5000 feet

metallic sulphides. Microscopic study of the cores at two to three inches below the surface showed the presence of large amounts of molluscan shell and up to several thousand foram shells per inch of core. At four to five inches the number of forams was less, indicating that there had been a concentration of forams in the muds near the surface of the core, possibly accomplished by a removal of fine silt by turbulence of wave action.

Cores were taken in the area between the nearest canal and leases 9861 and 9864. The locations of these cores are shown on the map, Fig. 6. There were six of these in an approximately straight line, about midway between the canal to wells 28 and 29 and the leases. The location of the line of cores was selected where the current velocity was least, as indicated by the great width of the bay at that point. Since tidal movement on ebb tide, as indicated previously, runs from northeast to southwest, when not diverted by wind, the cross-sectional width of the bay at the line of cores was more than two miles and the cross-sectional area close to 50,000 square feet. The flow from Rattlesnake Bayou into the bay (see Table 2) was measured by Lynch at about a maximum of 492.5 million cubic feet per 24 hour period providing a tidal current of less than 0.1 knot at the location of these cores. Wind drift currents may be slightly more than this ordinarily. If any part of the bay is subject to more deposition by reason of slow current, this area of bottom is most likely to accumulate either drifting spoil or silt. Any shoreward area is more likely to be scoured from wave wash than is the deep area selected, a half mile from the nearest shore.

These six cores were carefully studied both macroscopically and microscopically for evidence of recent silting. In Fig. 20 is a composite diagram of the six cores, giving both microscopic features and macroscopic characteristics. In Table 11 is a summary of the microscopic analyses made.

The cores showed that no silt had accumulated on the bottom in that area. There was nothing to indicate that the bottom was abnormal in any way. There was about a one-half-inch layer of loose aerated mud at the surface which contained shell and numerous

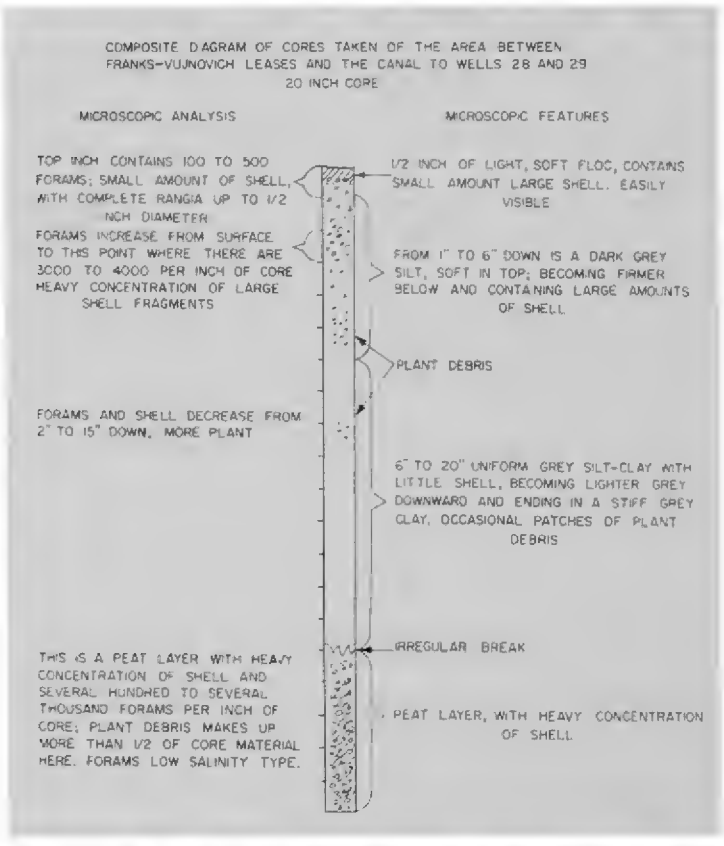
COMPOSITE DIAGRAM OF CORES TAKEN OF THE AREA BETWEEN
FRANKS-VUJNOVICH LEASES AND THE CANAL TO WELLS 28 AND 29
20 INCH CORE
MICROSCOPIC ANALYSIS
MICROSCOPIC FEATURES
TOP INCH CONTAINS 100 TO 500 FORAMS; SMALL AMOUNT OF SHELL, WITH COMPLETE RANGIA UP TO 1/2 INCH DIAMETER
1/2 INCH OF LIGHT, SOFT FLOC, CONTAINS SMALL AMOUNT LARGE SHELL. EASILY VISIBLE
FORAMS INCREASE FROM SURFACE TO THIS POINT WHERE THERE ARE 3000 TO 4000 PER INCH OF CORE HEAVY CONCENTRATION OF LARGE SHELL FRAGMENTS
FROM 1" TO 6" DOWN IS A DARK GREY SILT, SOFT IN TOP; BECOMING FIRMER BELOW AND CONTAINING LARGE AMOUNTS OF SHELL
PLANT DEBRIS
FORAMS AND SHELL DECREASE FROM 2" TO 15" DOWN, MORE PLANT
6" TO 20" UNIFORM GREY SILT-CLAY WITH LITTLE SHELL, BECOMING LIGHTER GREY DOWNWARD AND ENDING IN A STIFF GREY CLAY, OCCASIONAL PATCHES OF PLANT DEBRIS
IRREGULAR BREAK
THIS IS A PEAT LAYER WITH HEAVY CONCENTRATION OF SHELL AND SEVERAL HUNDRED TO SEVERAL THOUSAND FORAMS PER INCH OF CORE; PLANT DEBRIS MAKES UP MORE THAN 1/2 OF CORE MATERIAL HERE. FORAMS LOW SALINITY TYPE.
PEAT LAYER, WITH HEAVY CONCENTRATION OF SHELL

FIG. 20. Diagram of composite core from the bottom area of Lake Grande Ecaille between the canal to wells 28 and 29 and the Franks-Vujnovich leases.

FIG. 21. Fragments of shell of the Zebra Nerite from spoil in Lake Grande Ecaille. Spoil was from the canal to wells 28 and 29.

forams. Numbers of foraminiferan shells increased sharply from the surface downward and at two to three inches below the surface there were up to 4000 per inch of core. This large number could only be arrived at by a very long period during which the bottom was static, or by concentration due to a winnowing process. This latter is thought to be the explanation, for a static bottom has probably not occurred in the Mississippi River Delta where subsidence is so rapid.

CORES FROM THE AREA OF THE SPOIL FROM THE CANAL

Spoil from the canal to wells 28 and 29 was spread on the bottom in a broad band about 300 feet wide. All spoil was placed south of the canal. The spoil was not allowed to decrease the depth of the bay by more than six inches. Shifting of the discharge pipe, the end of which was mounted on a small barge, resulted in a large number of flattened piles, with intervening clear bottom. Six cores were taken in the area of the spoil and four of these passed through spoil, while the other two apparently missed the spoil, or passed through only thin sections.

The four cores containing spoil were studied to determine the characteristics of the materials removed from the lake bottom. All cores showed heavy concentration of shell in the top inch, decreasing in amount downward to the bottom of the spoil layer. The shell was of fixed marine and fresh-water (or near fresh-water) origin. Study of the shell fragments showed several fresh-to-brackish-water snails. One of these was vividly marked with black stripes on a white background, and was so abundant that it served as a marker for identifying the spoil (Fig. 21). This snail was the Zebra Nerite, *Purpurita pupa* Linne. According to Abbott (1954) this small snail is common in the West

TABLE 11

Data from microscopic analysis of cores from the area about midway between canal to wells 28 and 29 and the Franks-Vujnovich leases

Core no.	Core level	Forams	Shell	Remarks
193	Top inch	150–200	Small amount	Mostly fine silt with numerous forams
	2″ to 3″ from top	1000–2000	Large amount	
	4″ to 5″ from top	800–900	Moderate amount	
114	Top inch	75–125	Very small amount	Fine silt
	2″ to 3″ from top	2000–4000	Large amount	
	4″ to 5″ from top	75–125	Very small amount	
721	Top inch	300–400	Moderate amount	A live Tellina in surface (Mollusca)
	2″ to 3″ from top	3000–4000	Moderate amount	
	4″ to 5″ from surface	2000–3000	Large amount	
102	Top inch	100–200	Small amount	Several complete shells of Rangia in the surface
	1″ to 2″ from surface	100–200	Large amount	
	3″ to 4″ from surface	300–400	Moderate amount	Some shell to ½″ diameter
	10″ to 11″ from surface	500–600	Small amount	
	19″ to 20″ from surface (bottom of core)	50–100	Large amount	Forams are of low salinity (sand grain type)
747	Top inch	300–400	Small amount	
	2″ to 3″ from surface	2000–3000	Moderate amount	
	4″ to 5″ from surface	2000–3000	Large amount	
	12″ to 13″ from surface	50–100	Small amount	
	18″ to 19″ from surface (bottom core)	1000–2000	Large amount	
766	Top inch	200–300	Moderate amount	Some large pieces of shell to ¾″ diameter
	2″ to 3″ from surface	3000–4000	Moderate amount	
	4″ to 5″ from surface	100–150	Small amount	Clay nodules in this sample

Indies, and occurs in Florida. So far as the records show, however, it exists in Louisiana only as a fossil. It apparently was picked up in the deeper layers of the canal by the dredge. In our experience it has never been found elsewhere in Louisiana.

Microscopic study showed that foraminiferan shells were present in the spoil but in comparatively small numbers, the maximum number in one inch of core being about 100 and the minimum number two. It is assumed that the small numbers may have been partially due to dilution of the overlying marine silts by the larger amount of spoil derived from fresh-water deposits.

Samples of mud from the spoil were dried and ignited to determine loss on ignition (Table 10). This loss was small (8.3 percent) and the range of values in 12 samples was 4.0 to 12.3 percent. It is believed that the low content was due to a winnowing process as the spoil was pumped out of the end of the spoil pipe. This process concentrated the shell and heavy spoil in piles under the outlet and carried part of the lighter plant debris outside of the area of direct deposit.

PRELIMINARY CORES IN SOUTH GRANDE ECAILLE

In November, 1955, a large number of cores were taken in the area of leases 9861 and 9864, and additionally on leases which were not alleged to be damaged (Fig. 22). It seemed reasonable to assume that if silting had occurred it would be generally dis-

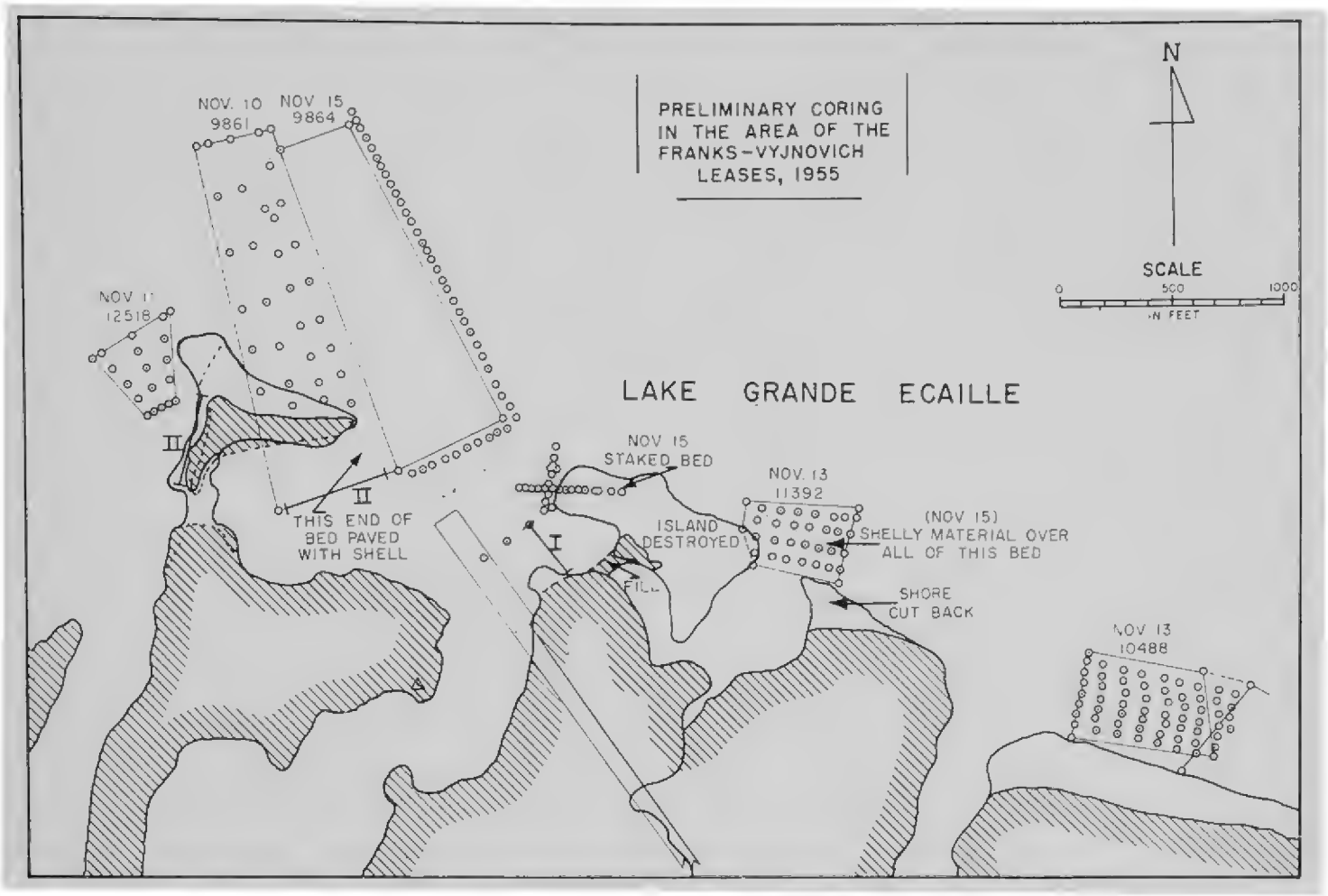

FIG. 22. Map showing locations of preliminary cores taken Nov. 10, 11, 13 and 15 on various oyster leases in southwest Lake Grande Ecaille.

tributed over the southern edge of the Lake. The coring showed that surface shell in some quantity was in all the areas investigated. The mud on these beds varied in consistency from mushy to stiff. All leases seemed "silted" in the sense that the mud was so soft that at least part of the area was questionable as good oyster bottom. However, the surface shell, and obvious shell distributed throghout the length of the cores, indicated that no part of the south part of the Lake had been recently silted. On the contrary, it was certain that the deposits of soft mud had accumulated over a long period of years.

Those preliminary cores which were taken on leases 9861 and around the perimeter of lease 9864 showed that there was much shell in the muds, both on the surface and for several inches down, the distance down varying with the core. Soft mud on these leases was more than a foot in depth in some areas. In other parts, where the artificial reef had been built up with *Rangia* shell, there was no silt at all. However, other parts of this artificial reef had been covered with silt up to several inches in depth. The mud on the built-up area contained shell fragments and forams as did other areas of the leases.

POSITIONED CORES ON LEASES 9861 AND 9864

The preliminary corings indicated that there was much soft mud on leases 9861 and 9864, but the shell fragments and foraminifera indicated that it had been there for a long time. Because these preliminary corings were only approximately located, it was decided to study the area further, and to concentrate the observations on those areas which the oystermen thought had been damaged. All corings were exactly located by two survey teams using standard surveying instruments. Some few of the earlier corings

had been thus located. Additional cores were taken on February 21 and on April 9, 1956. The locations of these cores were shown in the map, Fig. 14. All of these cores were analysed both macroscopically and microscopically, and many samples from the cores were analysed for comparative organic content by the ignition method. The following data are based on all of those cores which were exactly located by the Humble Oil and Refining Company engineers and plotted on Fig. 14.

The map shows (Fig. 14) that the system of coring was set up in the form of a series of lines beginning close to shore and progressing offshore. This pattern was adopted to test the theory that the soft silt on the leases originated on the eroding shorelines. This would be shown to be true if (1) the organic content was found to decrease significantly offshore, and (2) the foraminiferan content increased significantly offshore. Comparison of U. S. Coast and Geodetic Survey charts of about 1936, and of aerial maps of November, 1945, with a recent survey (1955) by the Humble Oil and Refining Company, showed that the island on which the Franks' camp stands had decreased by about one-half its original size in about twenty years. East Island, about five acres in size in 1935, had decreased in area to about one-half that size by 1945 and was a mere remnant by 1955 (see maps, Figs. 14 and 22). Thus a source of materials was evident.

Changes other than the pronounced shoreline erosion took place in the area of these leases. The most important of these was the breakthrough of the marsh strip lying between the small embayment to the south of Franks' Island and the pass from Lake Grande Ecaille to Bay Ronquille which lies just west of leases 9861 and 9864. It is not known when the marsh first gave way to form a channel to the south of Franks' Island, but the U. S. Coast and Geodetic Survey charts based on data taken in the early 1930's show the marsh strip to be intact. The aerial survey of 1945 shows the opening to be about 75 to 80 feet wide and the 1955 survey showed it had widened to about 150 feet. At some period between 1945 and 1955, attempts had been made to block the channel with shell, and at some time in the decade from 1945 to 1955 the opening had been about 225 to 250 feet wide. A strong scour current now passes through the cut and bottom in the cut is 10 to 12 feet deep.

Development of this passage was evidently gradual, as widening and deepening occurred. The destruction of East Island also contributed to diversion of flow through the cut. Some of the water, which formerly passed to the north of Franks' Island, was shunted to the south side, thus decreasing the current over the oyster beds. It is believed that this was one factor in causing the oyster beds to retain silt derived from the slumping shorelines.

Oyster Shell in the Cores. The exact location of the large number of cores taken in the area of leases 9861 and 9864 provided a means of determination of the extent of the area north of the island which was formerly used for oystering. Shell which has been long buried is characteristically grey to blackish in color due to deposition of metallic sulphides. Oyster shell may also be identified in other ways, even when fragmented. *Polydora* (an annelid worm) and *Cliona* (sponge) form characteristic borings in oyster shell, and *Membranipora* (an encrusting bryozoan) and barnacles attach to oyster shell. Laminations and sculpturing also are often characteristic.

These identification methods were used to determine (1) whether or not oyster shell was present in the mud of each core, (2) the depth to which it was found below the surface, and (3) whether or not it was found at the surface. These data showed that the area used for oyster production was formerly much larger than in 1956. The approxi-

mate boundaries of the area are shown in Fig. 15. It was indicated that nearly half of the formerly productive area has been abandoned. The abandoned area was to the south and east of the presently used parts of the leases. Along all of the east boundary of the abandoned section, the oyster shell was found seven to nine inches below the mud surface, extending upward for four or five inches, but not extending to the surface. Surface muds contained shell of other small molluscs but no oyster shell. The oyster shell was found at progressively higher levels westward and was at the surface 200 to 300 feet west of the eastern boundary. It was indicated that silting began in deeper water along the east boundary and progressively encroached to the westward. It is believed that the artificial reef was built because of these encroaching silts. The cores showed that the *Rangia* shell was placed on the silted base formed by a mound, the eroded base of which was formerly an island, a segment of the old bayou levee which ran in a north-south direction across what is now the west side of Lake Grande Ecaille. Sites of former islands are easily detectable by the peaty strata in cores.

Distribution of organic materials in the silts of the lease areas. The theory of the shore origin of silts was tested by determination of loss on ignition of mud samples from the cores. With exact knowledge of the location of each core and its distance from land, the relation of amount of loss of weight of dried mud on ignition, as a function of distance from shore, was worked out. The curve in Fig. 23 shows the decrease of ignitable materials with distance from shore. For each 100 feet of distance the mean decrease was 3.5 percent, certainly a significant figure. In Table 12 are presented figures on the loss on ignition of muds from cores at several stations around Lake Grande Ecaille, all from locations in marsh a few feet from the shoreline. These show the generally high organic content of the marsh muds, which are largely composed of plant remains.

The loss on ignition of muds on leases 9861 and 9864 was compared with that of (1) muds in the canal to wells 28 and 29, known to have been deposited from water-borne suspended matter, (2) muds from spoil removed from the canal and spread on the Lake bottom, and (3) undisturbed bottom between the canal and the oyster leases. The data are presented in Table 10. In the top six inches of mud there is little difference in the mean amount of the loss of ignition in the different areas, but the ranges of values differ significantly. Below six inches, where cores on the leases contained more of the peaty materials of the old levee, the values from the area of the leases and bay bottom were conspicuously higher than those of the spoil and the canal silts.

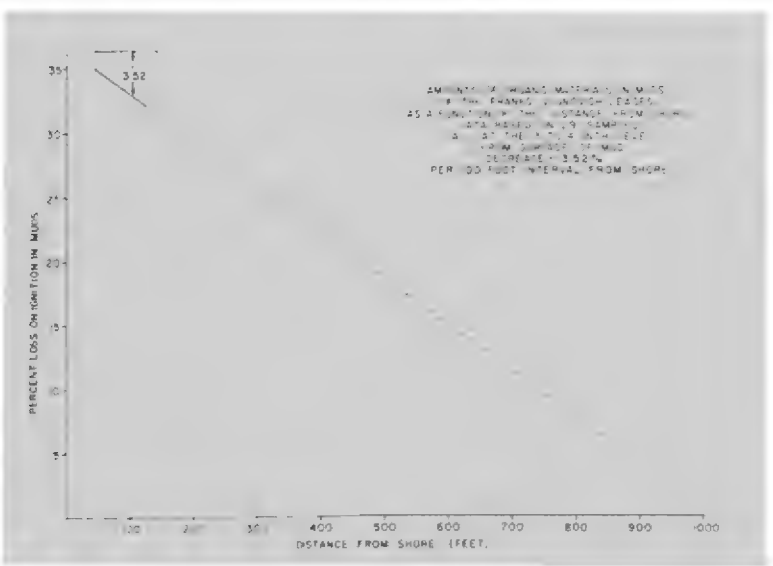

FIG. 23. Amounts of organic materials in muds on the Franks-Vujnovich leases as a function of distance from shore.

TABLE 12

Organic material in peat deposits: samples from parts of cores obviously made up of undisturbed plant debris around the shores of Lake Grande Ecaille

Core no.	Location	Core level distance from the top (inches)	Organic content; percent
16+2–21–56	Vujnovich lease	5–6	67.2
36+2–21–56	Pond on Franks' Island	8–9	78.2
25+2–21–56	Vujnovich lease	3–4	62.9
26+2–21–56	Vujnovich lease	5–6	63.6
33+2–21–56	Lease 10612 N. Grande Ecaille	6–7	59.5
34+2–21–56	Lease 10612	11–12	68.3
35+2–21–56	Lease 10612	6–7	70.1
1+3–23–56	Franks' Island in marsh	1–2	26.8
1+3–23–56	Franks' Island in marsh	6–7	70.1
1+3–23–56	Franks' Island in marsh	10–11	35.4
1+2–17–56	N. Grande Ecaille	12–13	58.6
4+2–17–56	N. Grande Ecaille	9–10	36.8
8+2–17–56	N. Grande Ecaille	11–12	59.4

Data on shell in the mud of the cores. Molluscan shell fragments were found in all silts of marine origin on the leases. Literally every core showed on macroscopic examination that shell, either in the form of entire valves or fragments, was distributed throughout the mud from the surface downward. Much of this shell was derived from *Rangia, Tellina,* hooked mussel, oyster or barnacles. Less abundantly other bivalves were represented. Some shell recognizable as belonging to the conch *(Thais)* was found, and borings by predaceous snails were common in *Rangia* shell.

Much of this shell material was in quite large pieces but ranged in size down to very fine fragments. Figs. 18 and 19 show samples from the coarsest sieves (1/10 and 1/20 inch mesh). The scale at the left of the dish is one inch.

It is not possible that the shell found in the mud on leases 9861 and 9864 was carried by the weak current of Lake Grande Ecaille over the intervening mile of bottom from the canals dredged by Humble Oil and Refining Company. Since there is no method of mixing the shell with silt except by natural accretion over a period of years as the silt accumulated, the presence of this shell showed that the silts were not a result of the dredgings.

The shell in the muds on leases 9861 and 9864 was carefully compared with that (1) in the silt accumulated in the canal to wells 28 and 29, (2) in the spoil dredged from that canal, and (3) from the intervening bay bottom. The silt in the canal contained only rare, very small bits (one exception of a fairly large fragment) of shell unrecognizable as to type, except that it was not oyster shell. The spoil removed from this canal had a high proportion of snail shells, including the above mentioned Zebra shell (Fig. 21) and no oyster shell. The shell from the intervening bay bottom was like that of the muds on the leases except that there was no shell of oysters, or of hooked mussels, bryozoa, or barnacles, which are commonly associated with oysters. Thus, even if it had been possible to transfer shell from the spoil to the leases, the difference in type of shell indicated that the fragments in the muds on the leases could not have come from the dredged canal.

Data from foraminiferan tests. Foraminifera also were distributed throughout the muds on the leases. Numbers found and levels in the cores shown in Tables 13 and 14. In Fig. 24 is shown the mean increase in numbers from the shoreline outward. The

increase in number offshore again indicated quite clearly that the origin of the silts on the leases was the adjacent shoreline.

The map, Fig. 15 gives the data on the depths of silt on the area claimed to have been damaged. It will be seen that most of the area was either not silted at all or had only insignificant silt. However, the north and west sides were covered with three to ten inches of mud. But the cores showed that most of this deep silt was not on the artificial reef at all. The boundary of this reef is roughly shown in Fig. 15. Only on the west side was the shelled area covered with more than one or two inches of soft mud. Incidentally the cores also showed that there was much old eroded oyster shell overlying the *Rangia* shell, indicating that the artificial shell reef had been built years ago. No area where *Rangia* shell had been recently spread was encountered in the coring operation.

Discussion of the data from the study of leases 9861 and 9864. The data accumulated in this study indicated that there was soft mud on the leases in certain places. But the data also showed that the silt had accumulated over a long period of years. The origin of some of the silt was indicated to be the disintegrating shoreline of the adjacent island (Franks' Island) and East Island. Silt also was undoubtedly accumulated from the generally distributed suspended materials always in the water. The beds became a depositing area, after being for a long time a scour area, because of the general subsidence of the bottom common to the entire region, and the opening and gradual increase in size of the cut south of Franks' Island. The presence of considerable molluscan shell and the normal foraminiferan shell accumulation in the muds showed that the deposition took place over a period of years rather than in the few months (November, 1955 to February, 1956) as claimed.

Of very considerable interest was the finding that the bay as a whole was not silted as shown by numerous corings along the shores and corings in the open bay. Even those cores taken quite close to canals indicated no silting other than that from direct deposition of spoil. It was indicated that the dredgings had changed nothing of a general ecological nature. No slowing of current occurred; if it had, silting would have followed. This indicates that spreading of silt over the bottom, raising the bottom by six inches (or less) has no measurable effect on wave scour of the bottom, even when the line of spread is at right angles to the prevailing current. The area of intense canal dredging in northeast Lake Grande Ecaille had no effect on the bottom elsewhere.

Depths over the beds were found to be greater than those shown on current navigation charts based on data at least twenty years old. This was a result of general subsidence in the area. It is believed that the build-up of silt on leases 9861 and 9864 was a result

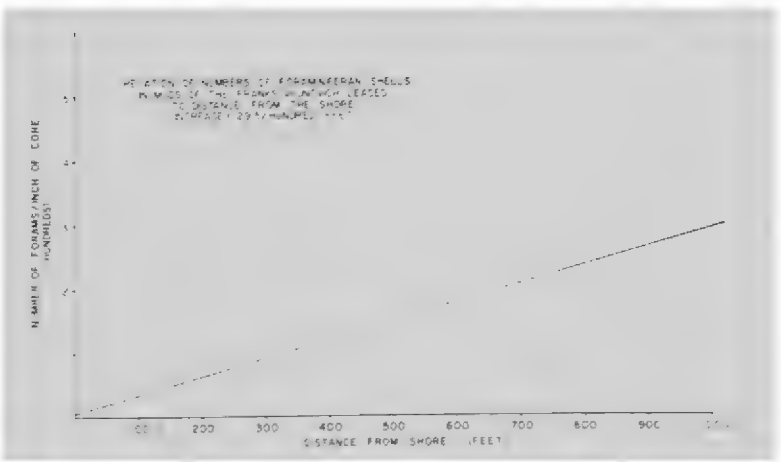

FIG. 24. Number of forams on the Franks-Vujnovich leases as a function of distance from the shore.

TABLE 13

Summary of foraminifera found in cores in the area of leases 9861 and 9864, Lake Grande Ecaille. Cores were taken on February 21, 1956

Core no.	Level: distance from surface of core 0–1″	2–3″	4–5″	6–7″	7–8″	8–9″	9–10″	10–11″	Depth to fresh-water deposit	Mean no. per inch of core
4	12	0	.....	.....	.....	.....	.....	.....	1–2″	6
5	23	7	.....	.....	.....	.....	.....	.....	2–3″	15
6	19	3	0	.....	.....	.....	.....	.....	3–4″	4
7	32	50–100	3	.....	.....	.....	.....	.....	4–5″	24
8	18	35	50	1	.....	.....	.....	.....	5–6″	26
9	150–200	300–400	28	0	.....	.....	.....	.....	5–6″	119
10	42	200–300	250–350	27	.....	0	.....	.....	7–8″	104
11	150–250	200–300	250–350	20	.....	8	.....	.....	8–9″	126
12	150–250	300–400	300–400	150–250	350–450	50–100	0	.....	9–10″	186
13	50–100	50–100	0	.....	.....	.....	.....	.....	3–4″	33
14	50–100	55	1	.....	.....	.....	.....	.....	3–4″	35
15	24	32	100–200	50–100	.....	5	.....	0	8–9″	35
16	100–150	30	20	.....	.....	.....	.....	.....	4–5″	50
17	150–200	50–100	200–300	9	.....	6	.....	0	6–7″	83
18	200–300	200–300	200–300	600–800	.....	15	.....	0	7–8″	202
19	200–300	100–200	15	3	.....	.....	.....	.....	4–5″	79
20	100–150	500–600	2	.....	.....	.....	.....	.....	3–4″	201
21	200–300	700–800	800–900	4	.....	.....	.....	.....	5–6″	426
22	50–100	150–250	1000–1500	0	.....	.....	.....	.....	4–5″	300
23	100–150	700–800	23	0	.....	.....	.....	.....	3–4″	206
24	250–350	300–400	400–500	.....	.....	10	.....	.....	8–9″	240
25	200–300	100–200	600–700	100–200	.....	5	.....	.....	8–9″	201
26	100–200	300–400	150–250	900–1000	.....	1200–1300	.....	300–400	12–13″	491
27	150–250	100–200	200–300	.....	.....	.....	.....	.....	Not found-short core	150

of this deepening, plus the diversion of current to the south of Franks' Island. The build-up of bottom was probably a consequence of these two factors.

Summary

1. Those characteristics of Louisiana bays and bayous which have to do with understanding of the problem of silting have been discussed. The levees of the Mississippi have had far-reaching effects on the ecology of the Mississippi Delta area by preventing new deposits of sand, silts and clays. The marshes have contributed unusual amounts of muds high in organic content, which make up an increasing proportion of bottom deposits in bays and bayous. Subsidence, compaction and rising sea level have produced conditions of high salinity and are responsible primarily for deepening and widening of bays and disintegration of shorelines. As sediments are transported to the sea and because little material is now deposited by the river, depths in bays are not in equilibrium with wind fetch and tidal current velocity. The amounts of turbid materials in waters of bays under natural conditions have been reviewed, as have conditions having to do with settlement of particles of clay, silt and sand on the bottom.

2. Conditions around working dredges have been studied. Measurements showed that silt is carried a maximum of about 1300 feet from working dredges under conditions for transport in shallow bays. Dilution has been shown to be effective in reduction of turbidities around working dredges. The amount of spoil drifting away from dredges was

TABLE 14

Foraminifera in the muds on the artificial reef area of the Franks-Vujnovich leases.
Cores taken April 9, 1956

Core no.	Order of distance from shore	Distance from shore	Forams in top inch of core
150	1	250	15
155	2	280	20
151	3	318	75
58	4	375	100
156	5	387	47
57	6	398	15
64	7	412	150
161	8	420	100
56	9	438	75
152	10	448	75
55	11	490	200
63	12	490	56
157	13	492	12
160	14	526	38
54	15	547	350
62	16	555	75
153	17	562	150
53	18	585	18
67	19	590	75
158	20	612	150
61	21	612	75
52	22	639	250
154	23	657	75
60	24	677	150
66	25	688	150
159	26	688	175
51	27	700	35
59	28	737	48
50	29	771	150
65	30	794	150

shown to be only a maximum of one percent of the total spoil dug from the bottom. Measurements of turbidities around working dredges have shodwn that in the close vicinity of the dredges these may be much higher than natural turbidities. At distances of a few hundred feet turbidities do not exceed those attained at times under natural conditions.

3. Experimental tests of the effect of suspended materials in water showed that concentrations up to 700 parts per million had little effect on oysters, either on mortality rate or on feeding.

4. The possibility that spoils might significantly reduce oxygen was carefully considered and was found to be unlikely under conditions found on normal oyster beds. It was also shown that the concept that dead oysters were significantly more effective in reduction of oxygen than were live oysters has no foundation in fact.

5. A concrete example of a problem involving claims of silting of oyster beds by canal dredging was thoroughly studied and the results of the study were presented. It was shown that extensive dredging operations by the Humble Oil and Refining Company failed to produce general silting in Lake Grande Ecaille. It was also shown that these dredgings had nothing to do with accumulations of silt on oyster beds in southwest Lake Grande Ecaille as claimed. The methods and procedures involved in resolution of problems of this type were presented. These involved analysis of many bottom corings at various points in Lake Grande Ecaille and on the oyster beds in question. It was

shown by these cores that the silt on the beds was derived from the disintegration of nearby shorelines and that the accumulation occurred over a long period of years.

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